REPRODUCTIVE MORPHOLOGY AND GENETICS OF *CARICA PAPAYA* (L.)

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ABSTRACT

This study was undertaken to examine the effects of climate and inheritance on carpellody and sterility in Carica papaya (L.). Sex in papaya is determined by a triple allelic series, "M_1m", "M_2m", and "mm", representing males, hermaphrodites and females, respectively. All combinations of the dominants are lethal. The hermaphrodite may tend toward femaleness (carpellody) or toward maleness (female sterility). Genetics for the factors carpellody and sterility have not been determined but it has been proposed that 2 different sets of factors are responsible. Changes in temperature and rainfall are known to influence the sexual expression of hermaphroditic plants but their specific degree of influence on several different known sexual forms of hermaphroditic plants have not been reported. Information about the climatic influences on sexuality in papaya will be useful in improving the commercial crop through breeding methods designed to minimize the effects of sexual changes. Three inbred hermaphroditic lines selected for highly stable expression of high carpellody, high sterility, and mostly normal elongata flowers were used.

Studies of floral initiation and development indicated that floral primordia were laid down 7 to 10 weeks prior to anthesis at the rate of about one new bud every 3 days. Differentiation of stamens began 7 to 8 weeks prior to anthesis and was completed by 5 weeks before anthesis. Ovaries began differentiating 6 to 7 weeks prior to anthesis and were complete by 4 weeks before anthesis.

The 3 inbred lines and their crosses (F_1, F_2, BC) were planted in 5 randomized incomplete blocks at Malama-Ki on the island of Hawaii. Sexual changes of all plants in the orchard were correlated to temperature and rainfall changes from 4 to 10 weeks prior to anthesis for a period of 1 year. Results indicated that sex response to climatic influence was greatest at about 7 to 8 weeks prior to anthesis.

Variations in sex expression from strongly carpellodic to strongly female sterile in the hermaphroditic trees were categorized into 7 "magnitudes" of expression. Trees were placed into one of these magnitudes in accordance with the sex expression of the last open flower each week and then placed into "classes" based on the overall yearly sex expression pattern of the tree. Results indicated that plants of the highly stable parents required as much as a 5°F. change in temperature before a shift in sex expression occurred. Plants with sex expressions lying between the parental types required smaller temperature changes for sexual shifts to occur. The response to changes in temperature appeared to be additive.

The segregation patterns of carpellody and sterility are also presented. The inheritance appears to be quantitative with partial dominance of carpellody. A genetic model is presented which utilizes a basic tri-hybrid ratio with 2 factors for carpellody (c/c, c/c) and 1 for sterility (s/s) and their normal alleles (+). "s" alleles were found to be epistatic over c alleles when "c" alleles were heterozygous. However, if "c" alleles were homozygous, carpellody would be exhibited regardless of "s". Expected and observed ratios were compared on this basis and chi-square indicated a close fit.

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INTRODUCTION

The papaya (Carica papaya L.) is indigenous to tropical America (22, 66, 78, 96). This tropical fruit spread rapidly along the early trade routes and by the 1900's, it was established in nearly all the tropical regions of the world (16, 22, 103). Its large melon-like fruit is produced almost continuously for several years. In Hawaii, papaya has become the most extensively marketed tropical fruit, next to the pineapple.

In spite of its popularity, many of the plant characteristics of interest to growers and researchers require further studies for greater understanding. Foremost of these is the formation of deformed fruits caused by the transformation of stamens into carpel-like structures (carpellody) under certain conditions and the loss of ovaries, causing female sterility, under other conditions. The former condition causes the production of unmarketable fruits while the latter condition prevents fruit production. It would be of interest to growers and plant breeders, not only to understand the causes of these problems, but also to control these effects through breeding and culture.

The purpose of this dissertation is to present new information about the sexual modifications of the hermaphroditic race of Carica papaya L., the inheritance of these modifications, and their behavior in relation to environmental conditions.

LITERATURE REVIEW

A. BOTANY AND SEX OF THE PLANT

Caricaceae is a small family in the order Parietales, consisting of 4 genera of which Carica is the most important. There have been about 60 species of Carica reported (19, 34, 50, 56, 66, 67, 78, 81, 96, 101, 108, 109, 110, 112, 113, 144). However, a more recent work by Badillo (9, 10, 11, 12) has reduced this number to approximately 30 by identifying duplications and synonymy.

The family Caricaceae has been shifted taxonomically several times. Hooker (46) placed it near Passifloraceae as an anomalous tribe. Later, Usteri (108) and Wordsell (114) placed it near Euphorbiaceae. Then, Van Tiegham (109) placed it near Cucurbitaceae. Others (24, 81, 110, 112) have described it as a separate family related to Passifloraceae. It is generally agreed today that Caricaceae is a separate family related to both Passifloraceae and Cucurbitaceae (19).

The papaya (Carica papaya L.) is a tall herbaceous plant with heights ranging up to 30 feet. The plant usually consists of a central hollow stem topped by a tuft of large lobed leaves borne on the end of long petioles. Generally, only a single stem is formed, but pinching the terminal bud or cutting back (called stumping in Hawaii) a fully grown tree may produce a tree with several secondary stems formed from the central stem (3, 69).

Leaves are generally produced in a regular phylotaxy (2/3, 3/5) and when removed from the plant, leave a large leaf scar (3). These scars may be nearly triangular at the top of the plant, but towards the bottom

their vertical axis is reduced while the horizontal axis is extended until the scar appears as a thick line running half way around the stem. The distortion of the leaf scar may be due to lateral strain since C. papaya is not an entirely woody plant (3).

The stem is composed mostly of parenchyma tissue which remains meristematic for the life of the plant. This parenchyma is interspersed with scattered xylem tissue and no interfascicular cambium is formed. There is a meshwork of sclerenchyma tissue exterior to the wood cylinder. This is a rather heavily lignified tissue but apparently not heavy enough to support the entire plant and, therefore, the plant relies mainly on turgor for rigidity (3).

Early descriptions of papaya identify the trees as being dioecious (66, 67, 75). However, other forms having perfect flowers were noted and one worker (15) described a form having hermaphroditic flowers as a new species, Carica hermaphroditica. It soon became apparent that many sex forms of C. papaya exist and several workers undertook to describe them. Higgins and Holt (34) describe more than a dozen individual forms and this is probably the first attempt to catalog them. Higgins (35) lists seven of these as being the most important. In addition to the ordinary staminate (male) and pistillate (female) trees, he lists the correae, elongata, sterile hermaphrodite, forbesi and pentandria. This and studies conducted elsewhere finally resulted in the description of C. papaya as a polygamous species (100).

Hofmeyr (36, 37) and Storey (97, 98) working independently in South Africa and Hawaii, respectively, came to the same conclusions with respect to the inheritance of the three basic sex forms in Carica papaya.

These three sex forms are recognized to be staminate (male), pistillate (female), and hermaphrodite. Storey (97) revised earlier lists to include five important forms; one pistillate, three hermaphrodites and one staminate which he labels types I, II, III, IV, and V, respectively. Storey (97) also reported that others have described these five forms (14, 30, 34, 57). Hofmeyr (36) listed only the three basic forms. In the reports of Hofmeyr (36, 37) and Storey (97, 98) the sex forms noted as II, III, and IV were taken to be genetically alike; i.e., hermaphroditic. Various workers (34, 69, 97) have noted the formation of some fruit on the long flowering racemes which characterize the staminate tree. This tree is still referred to as a male, however. Hamilton and Izuno (31) have offered the hypothesis that the long raceme characteristic of the male tree is only a sex linked secondary character and that a tree which produces bisexual flowers on these racemes is actually a hermaphroditic tree and one that produces female flowers is a female tree. It was further suggested that the classical concept of three genetic types advanced by Storey and Hofmeyr may have to be revised in view of this new hypothesis to include only two basic sex types. For the present, however, the classical concept to be described below will be followed.

Hofmeyr (36, 37, 38, 39) and Storey (97) reported, after determining the ratios of sex forms obtained from controlled crosses between the sex forms, that sex in the papaya is determined by a single gene with three alleles. Hofmeyr (36) assigned the symbols M_1 to the staminate, M_2 to the hermaphrodite, and m to the female (pistillate). Storey (98) reported all combinations of the dominants M_1M_1, M_2M_2, and M_1M_2 to be lethal to the zygotes. Hofmeyr's conclusions (36) were that males and

hermaphrodites are enforced sex heterozygotes and therefore, the genotypes are: M_1m, male; M_2m, hermaphrodite; and mm, female. Storey (98) supported this conclusion in a later report. Elden (21) and Kumar, et al. (59) regarded sex determination in species where males and hermaphrodites are heterozygous and females homozygous as being of the XY type.

Hofmeyr (42) postulated a genic balance hypothesis to explain sexual stability or instability among the sex forms. Using this method, the genotype mm has a value of 3 in favor of femaleness. M_1m has a value of 1 in favor of maleness and M_2m has a threshold value of 0. He believed that proximity to the threshold value is reflected by the sexual instability of males and hermaphrodites which show rather pronounced variation in sexual expression, whereas the relatively high value for femaleness is indicative of the stability of the female tree.

Narasimham (73) acknowledged the 13 morphological forms of Higgins and Holt but stated that several of these forms are genetically identical but differ functionally by the influence of modifying genetic factors. He agreed with Hofmeyr's and Storey's three basic sex forms.

Kumar (61) listed 6 sex types. Five of these are the same as described by Storey (97) and Wolfe and Lynch (113). The other one is called a type IV+ and is inserted in the lineup between types IV and V. Storey (97, 100) labeled this type as being a "summer sterile" hermaphrodite. Kumar (61) stated that it is found on hermaphroditic trees as a result of certain environmental conditions and is functionally staminate. Kumar also described the occasional co-existence of various flower types on a single tree.

Storey (99) reported that sex in papaya may be determined by a complex of genes instead of a single gene. He stated that these genes probably lie closely linked in different segments occupying identical regions on the sex chromosomes. Hofmeyr (44) also suggested that modifying genes might be responsible for sex reversal in staminate and pistillate trees. He also stated that different sets of modifying genes are responsible for the sex changes in flowers of the opposite sexes and further postulated a complex genotype for these factors. Sex reversing staminate trees were noted by many workers (19, 34, 51, 97). However, the only case of a sex refersing female tree noted in literature was by Hofmeyr (38, 39). He stated that after several generations of inbreeding, he was able to obtain some stamens on the female trees in some cultivars but not in others.

Singh, et al. (92) agreed with Storey and Hofmeyr in the presumption that sex reversal in papaya may be due to an interaction of genetic and environmental factors with modifying genes also participating. They also stated that there are probably two types of "male" trees, the fruiting type which undergoes sex changes and the non-fruiting type which produces only staminate flowers. They believed the genetic makeup of these two types to be different.

Storey (100) subdivided his three basic sex forms into eight general categories designed to include all of the almost innumerable intergrades of sex into easily recognized groups. He assigned the following names: staminate, teratological staminate, reduced elongata, elongata, pentandria, carpelloid pentandria, and pistillate. Storey (100) also advanced his new hypothesis that there are two sets of genetic factors which modify

sex expression of flowers in male and hermaphroditic trees. It had been previously stated that the primary sex-determining factors may be identical in the male and hermaphroditic trees (36, 47, 54, 99). One set of genetic factors would be responsible for seasonal shifts from female fertility to sterility and back to fertility by affecting carpel development and the other for causing a change in stamen morphology, i.e., from normal stamen structure to a carpellodic structure with fusion to the original pistil and back again. Based on year-round observations of flower form, Storey (100) listed what he terms as 32 heritable tree sex forms.

Lassoudiere (64) in a recent paper presented a botanical description of the various types of inflorescences observed by him. These descriptions follow closely those already given.

In order to produce marketable fruits, an orchard must be made up of female trees in some areas of the world where the female fruit is preferred and of hermaphroditic trees in areas of the world where the pyriform or elongata type of fruit is preferred. In dioecious lines, half of the population planted in the orchard would be males and unproductive. Therefore, it is desirable to be able to classify the sex of young papaya seedlings before field transplanting. Reyes (83) reported that sex of young papaya seedlings could not be determined by any known methods or suspected indicators such as the margin of initial leaves, color and vigor of stem, or size and extent of root systems. He also stated that decapitation is not a reliable method for causing sex reversal.

Sakarai (84) reported negative results between seed size and sex and external characters of seedlings. However, he reported that seeds

producing female plants are slightly heavier than those giving rise to staminate plants. Pope (77) also reported negative results in trying to segregate seeds into "he's" and "she's". In what is probably the only record of positive results, Barnes (13) stated that the more strongly growing plants in the nursery can be weeded out as being invariably males. This had been previously contradicted by Reyes (83) and Sakarai (84). Hofmeyr (36) reported on extensive experiments on the relation between vigor and sex in seedlings and concluded that there is no difference in vigor between male and female plants in the first year (before fruiting). He concluded, however, after several seasons, that the staminate plants become progressively more vigorous in comparison to pistillate plants. This may be due to production of fruit on females and the lack of it on males.

Kumar (60) concluded that perhaps seed of a deeper brown color produces a higher percentage of fruit bearing plants. He also suggested that at the age of 12 weeks, the seedlings which later bear staminate flowers show a higher rate of stem elongation than those of pistillate or hermaphroditic classes.

Singh, et al. (93) attempted to identify seedling sex at the nursery stage by use of colorimetric tests. They reported moderate results in that 67 percent and 87 percent of the seedlings classified respectively as male and female prior to flowering were classed correctly.

As indicated by the results reported by these workers, attempts to sex young plants prior to flowering have been largely unsuccessful. Therefore, several seeds must still be planted in each hill and at flowering time a vigorous and productive female or hermaphroditic plant

is selected to grow with the others rogued out.

B. FLORAL MORPHOLOGY

One of the earliest and most complete descriptions of the papaya flower was undertaken by Mello and Spruce (69) from plants observed in Brazil. Flowers were described as arising in axillary racemes on panicles "whereon the secondary peduncles are cymuliferous, the axial flower being more precocious than the rest, and often diverse in structure." The flowers were described as being usually unisexual and dioecious "but sometimes polygamous, with flowers all female on some plants, all male on others, and on others bisexual and male."

Their calyx is free, minute and 5 lobed. The corolla is composed of 5 ligulate petals which are more or less united into a monopetalous corolla but in female flowers, free to the base. Stamens were described as being 10 in two rows fixed on the throat of the corolla by very short filaments. Anthers are erect and split lengthwise. The female ovary is free, one-celled with 5 parietal placentae. Stigmata are found to be 5 lobed, continuous with the carpels and alternate with the petals.

Mello and Spruce (69) described more fully the three basic forms of the flower. They described one form (Form one) as being female with its inflorescence axillary, peduncles solitary, 1 or 2 inches long and bearing 2 to 4 shortly pedicellate flowers. They reported the terminal flower to be quite precocious and as a result, all other flowers on that peduncle fall away unfertilized. The corolla consists of 5 distinct petals. Stamens and staminoidia are completely absent. The ovary is

ellipsoid, one-celled with 5 nearly flat parietal placentae. The style is continuous with the ovary, very short with "5 linear, fleshy, flattened, radiating, dilated, and subpalmately laciniated" stigmata at the apex. They are stigmatose on the whole upper surface.

Form two was described as the bisexual and male form. The peduncles, again, are axillary and divided into 3 at the base. The lateral branches are 2 to 8 inches long with 1 or only a few flowers. The medial branch is 12 to 24 inches long. Fruits are borne somewhere above the middle 2 or 3 partial peduncles which are further subdivided into 4 or 5 flowered pedicels. There are from 1 to 4 bisexual or fertile flowers on each panicle. The corolla is gamopetalous and 5 lobed. There are 10 stamens inserted in the throat of the corolla in two series. The ovary is oblong and almost club shaped. Stigmata are about the same as in the female ovary but slightly smaller. In the male or sterile flowers the corolla is gamopetalous and 5 lobed. Stamens are as in the bisexual flower but the pistil is rudimentary, short and tapering upwards.

Form three was described as a male flower differing from Form two in that ovaries are absent. The terminal bisexual flower is uniformly abortive or absent.

The above descriptions by Mello and Spruce (69) are quite complete and accurate in their details of the three basic forms of flowers. However, Higgins and Holt (34) and Higgins (35) described tree forms based on observations of flower types and subsequently listed morphological descriptions of more than a dozen types ranging from male to female.

Haigh and Fernando (30) in a planting experiment to determine the number of seedlings needed per "hole" to assure a productive (female) orchard, noted five different types of flowers being formed and labelled them types A, B, C, D, and E, respectively. Type A is a typical male flower with petals free for only 1/3 their length, 10 stamens, a rudimentary ovary, a short style, and no stigma. The flowers are borne in numerous clusters on long branched pendulous stems. Type B is a male hermaphrodite, larger and broader than type A with petals free for 1/2 their length, 10 stamens, a rudimentary ovary, a short style, and no stigma. These flowers are borne in clusters of about five on short stems. Type C is a hermaphrodite. It has petals free for about 3/4 their length, 10 stamens, an elongated ovary with 5-lobed stigma and produces elongated fruits. Flowers are borne in small clusters of 1 to 3 on short stems. Type D is a female hermaphrodite. Petals are free for their entire length. There are 5 stamens and a 5-lobed stigma. Flowers are borne in clusters of about 3 on short stalks but only one matures. Type E is a typical female with petals free for their entire length and a conical flower bud. There are no stamens. The ovary is 5-lobed, ovoid and has a 5-lobed stigma. Flowers are borne singly or in clusters of 3 on very short stalks.

Storey (97) described the flowers of the same 5 sex types as previous workers, but noted that in transitional types such as his Type III, the number of stamens is variable from 2 to 10 and that their degree of adnation to the pistil or corolla also varies. He also noted that a high degree of pistilloidy may occur among the stamens and that the pistilloid structures could appear in any stage of development. The

pistils themselves may also be distorted with fusion of carpels highly variable. In his Type IV which he called elongata, the petals are gamopetalous for 3/4 of their length and free from the ovary except at the very base. The 10 stamens are arranged in the throat of the corolla tube in 2 whorls of 5 each. One of these whorls is opposite the corolla lobes and subsessile while the other is alternate with slightly longer filaments. All 10 filaments are adnate to the corolla tube to its base. The pistil is generally 5 carpellate but can range from 1 to 10.

Devi (19) presented a rather complete description of the vascular anatomy of the hermaphroditic papaya flower. He traced the entire vascular system as each set of bundles split off from the parent stele to enter the various parts of the flower from its bracts to its stigmatic lobes.

Horovitz (48) described the sexual variability of the flowers of andromonoecious and androecious papaya plants. He stated that a good evaluation of flower sex can be obtained from measurements of the ratio of length to width of the fused portion of the corolla and the ratio of the length of the fused portion to the free portion. Kulkarni (58) also reported on sex changes in this type of papaya plant.

Storey (101) presented a theory which deals with the derivations of the unisexual flowers (monoecious and dioecious) of C. papaya. He postulated that the flowers of C. papaya evolved intraspecifically along two lines of divergence from the hermaphroditic elongata type and that each line culminated in a type which is unisexual both structurally and functionally. Parkin (76) also presented evidence which suggests that all unisexual angiosperm flowers are derived from hermaphroditic

ancestors. Storey (101) suggested that staminate flowers were derived in the classical manner, i.e., by phylogenetic reduction which led to the loss of the gynoecium. The pistillate flower, however, was derived from hermaphroditic flowers in which stamens became carpel-like and suppressed the original carpel in two series. Anatomical evidences were shown which tend to substantiate these theories. In a later paper, Storey (102) discussed further the concept of the derivation of the pistillate flower with special emphasis on the vascular traces of that flower.

C. CYTOLOGY

The earliest observations on cytology of Carica papaya were made by Usteri (108) and Kratzer (56). They both studied development of the embryo sac. Usteri reported that the micropylar cell of the tetrad gives rise to the embryo sac, but Kratzer noted that any cell of the tetrad may give rise to the embryo sac. Heilborn (32) also reported on early development of the embryo sac confirming earlier results obtained by Kratzer. Agharkar and Banerji (1) reported that the chalazal cell of a linear tetrad of megaspores gives rise to the mature 8 nucleate embryo sac in Carica papaya.

Heilborn (32) was among the first to study the chromosomes of C. papaya. In all cases he reported the diploid number to be 18. Meurman (71) reported the presence of 9 gemini in the pollen mother cells of C. papaya and the absence of any sex chromosome pair in them. Sugiura (104) reported the same results and in addition, the formation of a

multipolar spindle. Lindsay (65) also noted that no evidence can be found for an unequal pair of chromosomes in the division of the pollen mother cells. Asana and Sutaria (4) confirmed earlier reports of a meiotic number of 9 chromosomes and reported multipolar and tripolar spindles. Foster (25) in a very complete investigation of the cytology of C. papaya listed several observations tending to confirm most other reports of early development and a haploid chromosome number of 9. Hofmeyr (36, 37) and Storey (98) independently reached identical conclusions that a heteromorphic pair of chromosomes is not to be found in C. papaya. Kumar, et al. (59) reported on a comparative cytological study of the Carica species and 21 regional types of C. papaya collected from various parts of the tropics. In all cases they reported the chromosome numbers to be n = 9, 2n = 18. They also reported that there are no differences in the morphology of the chromosomes in the hermaphrodite, female, and male sex forms of the same variety. They observed in the male papaya, however, that one particular bivalent showed precocious anaphasic separation. They suggested that this phenomenon is in keeping with the expected behavior of a heteromorphic pair. Storey (99) also noted this. Kumar, et al. (59) noted from their study of the somatic chromosomes that there are no detectable differences between the chromosome complement of one sex form and another. Their final conclusion was that variation in sex expression in C. papaya is not associated with any visible morphological difference between the autosomes and the sex chromosomes. Van Elden (21) also reported no heteromorphism in a chromosome pair.

Hofmeyr (43) suggested that cytogenetic studies carried out by him on male and female sex forms of C. papaya indicated the possibility that the sex ratio may be controlled using trisomics so that from 66 per cent to 100 per cent of the progeny of certain crosses would be composed of female or fruit bearing trees. This has not been realized as yet.

Storey (99) reported that crossing over occurs freely and that this freedom of crossing over lends support to the hypothesis that the sex determining factors are confined to small differential regions on the sex chromosome within which crossing over is precluded.

D. EFFECTS OF INTERNAL AND EXTERNAL ENVIRONMENT ON SEXUALITY

At present, there is very little information available concerning the internal factors which may influence the sexuality of the flowers produced on any given type of papaya tree. It is very difficult to gain such information as specific substances are difficult to isolate and their relationships to specific plant actions are not easy to prove. Use of presently available techniques allow, for the most part, only speculation as to specific genetic factors.

Horovitz (48) postulated that the occurence of sexual variability is dependent upon the presence of alleles "F" or "F^h" and that within a given genotype, temperature is the decisive factor for the type of flower produced at a given moment. Also, the existence of specific male and female florigenic substances was postulated.

Sayed (86) stated that the plants observed by him showed no particular stability and exhibited all the types listed by other workers;

also, that these plants appeared to be influenced by genetic factors, soil variation, and environmental conditions.

Genetic factors which affect sexuality have been investigated by several workers on many different plants (2, 18, 23, 26, 29, 33, 53, 74, 90, 91, 111, 115, 116, 118). Most of the results of these research reports were best summarized by Emerson (23): "Even the occasional appearance of sex intergrades approaching the condition of hermaphroditism may well be due to the influence of several heterozygous sex factors of relatively minor influence ... modifying factors ...."

Much more information is available concerning environmental effects. Various experiments have shown that growing conditions can have a profound effect upon the sexuality of flowering plants. Howlett (49) noted that tomato plants shift towards femaleness with a decrease in temperature. Quagliotti (80) showed a similar shift in the carrot. Schaffner (87) reported that hemp is so sensitive to any external influence that sex reversal can occur when the plant's metabolic level is disturbed or changed. The castor bean also shows sexual fluctuations which may be influenced by modifying factors in response to environmental conditions (18, 89, 90, 118). Flowers of the cucumber vary widely in the ratio of staminate to pistillate flowers and this is attributed, at least partially, to environmental influences, mainly temperature (20). Chemical applications of Gibberellin also alter sex of the cucumber (72) as well as many other plants (82).

Awada (5) reported that conditions of drought tend to suppress the gynoecium and more male flowers are produced in C. papaya. In another report, Awada and Ikeda (6) reported that high levels of nitrogen may

promote femaleness and depress male sex expression. Choudhori, et al. (17), however, stated that total carbohydrates and catalase activity are higher, while nitrogen is lower in female flowers as compared to male flowers. They further postulated that due to this physiological difference from femaleness to maleness, it may be possible to control the sex of some plants.

Awada (7) also reported that minimum temperatures appeared to influence sex expression by promoting female flower production while suppressing male sex expression. Awada (8) also noted that male sex expression may be promoted by defoliation whereas defloration may promote femaleness. In general, many workers agree that conditions of stress tend to promote maleness or inhibit the female portion of the flower, while generally good growing conditions promote femaleness (5, 6, 7, 8, 33, 62, 63). Many workers have also reported on the seasonal variations of sex type in papaya flowers. Most agree that the flowers shift to maleness after warm periods and femaleness following cooler periods (7, 27, 28, 55, 57, 62, 94). However, Teaotia and Singh (105) reported opposite results in plants subjected to seasonal temperature changes.

EXPERIMENTAL PROCEDURE

A. PLANTS USED IN STUDY

Hermaphroditic plants of several lines of Carica papaya L. from the breeding program in the Department of Horticulture, University of Hawaii were used in this study. Three inbred lines differing widely from each other in sex expression were selected. Line 13BF6 was selected for a high degree of female sterility, Line 17CF6 was selected for high production of carpellodic flowers throughout the year, and Line 17AF6 was selected for production of a high percentage of flowers which were "normal", i.e., of the elongata type described by Storey (97), throughout the year. All the lines were inbred for 6 generations, and are highly stable in the types of flowers produced under Waimanalo conditions.

After the fifth generation of inbreeding, crosses were made among the three inbred lines (Table I).

TABLE I. CROSSES MADE BETWEEN THREE INBRED LINES AND THE PROGENIES OBTAINED

P_1		P_2	F_1	F_2	BCP_1	BCP_2
17AF6	X	17CF6	X	X	0[a]	X[a]
17AF6	X	13BF6	X	X	X	0[a]
13BF6	X	17CF6	X	X	X	0[a]

[a]Seeds of these crosses did not germinate and plants were not available.

Two sets of plants were established. One set consisted of plants of the three inbred lines and was grown at the Manoa campus. Seeds of these lines were started in community pots and transplanted to jiffy pots at the cotyledonary leaf stage. They were then transplanted to 5 gallon containers when about 6 inches tall and grown in a soil mix composed of soil, volcanic ash, and peat moss in approximately equal proportions. The 5 gallon containers could be moved but were large enough to permit normal growth during the experiment.

The plants were divided into three groups at the initial flowering stage. Each group contained 5 plants each of Line 17CF6 (high carpellody), Line 17AF6 (highly normal), and Line 13BF6 (high sterile). One group was used as the check and was grown outdoors. The second group was placed in a glasshouse. Diurnal temperature variations within the structure ranged from a low of 70°F. to a high of 110°F. The low temperature was usually at least 5°F. higher than outdoors due to heat retention in the glass structure during the night and the high temperature was usually 15° to 20°F. higher than outdoors. The third group was grown outdoors under shade from sunrise to 9 A.M. and from 4 P.M. to sunset, under partial shade from 9 A.M. to 4 P.M., and was moved into a large reefer box at night. The temperature in the reefer box was held at 52°F.±2°. This temperature was selected because it would subject the plants to a non-injurious cold temperature 10° to 15°F. lower than the check. The shady periods during the day kept the temperature of group III 3° to 5°F. lower than the check which was grown in full sunlight.

The second set of plants consisted of the three inbred lines (sixth generation) and their crosses. The seeds were germinated in community pots, transplanted to jiffy pots when the first two leaves appeared, and transplanted to the field when about 6 inches tall. Two plants were planted per hill to increase the possibility of obtaining one hermaphroditic tree in each hill. The progenies and parental lines were planted for this study at Malama-Ki Experimental Farm on the island of Hawaii, the principal papaya producing region in the state. The mean yearly temperature is 73.5°F. with a range from 63°F. to 83°F. The average diurnal temperature variation is 13.5°F. The average yearly rainfall is approximately 143 inches (106), normally well-distributed although rainfall increases notably during the winter months from December through March and 1 to 3 months of drought can occasionally occur. However, rainfall during the year of this experiment was approximately 183 inches, some 40 inches above normal. The pattern of rainfall, however, was the same as for more normal years. Soil at Malama-Ki is of the volcanic type called 'aa'. A member of the lithosols group, it is composed primarily of porous lava with a limited amount of volcanic ash, weathered rock material, and some organic material (107).

Because of insufficient numbers in some progenies, 5 randomized incomplete blocks with 8 to 16 plants per progeny were used. Fertilization practice consisted of one pound of a 10 - 10 - 10 fertilizer per plant every 2 months.

B. ESTABLISHING CLASSIFICATION METHOD FOR FLOWERS AND TREES

Data on sex of all plants were taken as soon as the first flowers reached anthesis. Once a week, the sex type of the last flower opened was recorded and placed into one of the 7 possible Magnitudes of sex expression (Table II). The sex (Magnitude) of each type of plant was plotted on graphs to show the time and degree of sex shift during the 52 week period of data collection (Plate I, Figs. 1 - 11). Each tree was then placed into one of 11 different classes based on the overall yearly sex expression of the tree (Table III). Trees were classified both on the basis of the mean magnitude of the sex of the flowers and the amount of variability between magnitudes.

C. DESCRIPTION OF CLASSES

Each of the line graphs representing the sex variation in plants found in the 11 classes is a graphic representation of the typical plant that fell into that class (Plate I).

The plants of Class 1 (Plate I, Fig. 1) were highly carpellodic and varied in magnitude only from 1 to 2, the two highest levels of carpellodic sex expression. These plants were near the end point of sexual expression and did not have a great latitude for change except to become less carpellodic. Conditions at Malama-Ki apparently never varied enough to cause any shift of sex expression greater than to Magnitude 2. Plants remained at the highest level of carpellody (Magnitude 1) for more than 70 per cent of the year.

TABLE II. A DESCRIPTION OF THE HERMAPHRODITIC FLOWER TYPES OBSERVED

MAGNITUDE	DESCRIPTION
1.	A completely carpellodic flower. Female-like and with no stamens.
2.	A carpellodic flower with 1 to 4 stamens which may be in various stages of transformation or adnation to the ovary.
3.	A carpellodic flower with 5 normal stamens arising from the base of the ovary. Referred to as "Pentandria" by most workers and sometimes called a "normal" type flower.
4.	A carpellodic flower with 6 to 9 stamens. At least 5 of these are normal stamens while the remaining 4 may be in various stages of transformation or adnation to the ovary.
5.	A "normal" flower with 10 complete stamens and a 5 carpellate ovary. Male and female parts show no transformational stages and the ovary is smooth and pyriform. This flower is referred to as "elongata" by most workers.
6.	A flower with 10 normal stamens and a 1-4 carpellate ovary. It is moderately female sterile and is referred to as "reduced elongata" by most workers.
7.	A flower with 10 normal stamens and an ovary that is reduced to a vestigial spike. It is completely female sterile.

TABLE III. TREE CLASSES BASED ON OVERALL YEARLY SEX EXPRESSION

CLASS	DESCRIPTION
1.	Very high carpellody -- Trees were in Magnitude 1 more than 70 per cent of the time, in Magnitude 2 for the remainder of the year. Mean magnitude was 1.00 - 1.10.
2.	High carpellody -- Trees ranged from Magnitudes 1 to 4 and were in Magnitude 1 for less than 20 per cent of the year. Mean magnitude was 1.11 - 2.40.
3.	Carpellodic -- Trees were in Magnitudes 2 or 3 for 50 to 75 per cent of the time and in Magnitudes 4 or 5 for the remainder or spent less than 25 per cent of the year in Magnitude 5. Mean magnitude was 2.41 - 3.30.
4.	Slightly carpellodic -- Trees which were in Magnitudes 2 to 4 for about 70 per cent of the time and in Magnitude 5 for the remainder. Mean magnitude was 3.31 - 3.91.
5.	Very slight carpellody -- Trees were in Magnitude 5 for more than 70 per cent of the time and in Magnitudes 3 or 4 for the remainder. Mean magnitude was 3.92 - 4.64.
6.	Normal to very slightly carpellodic OR very slightly sterile -- Trees were in Magnitude 5 for more than 80 per cent of the time, and in Magnitudes 4 or 6 to 7 for the remainder, scattered throughout the year. Mean magnitude was 4.65 - 5.20.
7.	Very slightly sterile -- Trees were in Magnitude 5 for more than 70 per cent of the time and in Magnitudes 6 to 7 for the remainder. Mean magnitude was 5.21 - 5.56.
8.	Slightly sterile -- Trees were in Magnitude 5 about 50 per cent of the time, and in Magnitudes 6 to 7 for the remainder. Mean magnitude was 5.57 - 5.92.
9.	Sterile -- Trees were in Magnitudes 6 to 7 50 to 75 per cent of the time, and in Magnitudes 5 or 6 for the remainder, with less than 25 per cent in Magnitude 5. Mean magnitude was 5.93 - 6.28.
10.	High sterile -- Trees were in Magnitudes 6 or 7 more than 80 per cent of the time, in Magnitude 5 less than 20 per cent, and in Magnitude 7 less than 70 per cent. Mean magnitude was 6.29 - 6.70.
11.	Very high sterile -- Trees ranged from Magnitudes 6 to 7 and were in Magnitude 7 more than 70 per cent of the time. Mean magnitude was 6.71 - 7.00.

PLATE I

Variability in magnitude of classes 1 through 11 over a 52 week period.

Figure 1. Class 1

Figure 2. Class 2

Figure 3. Class 3

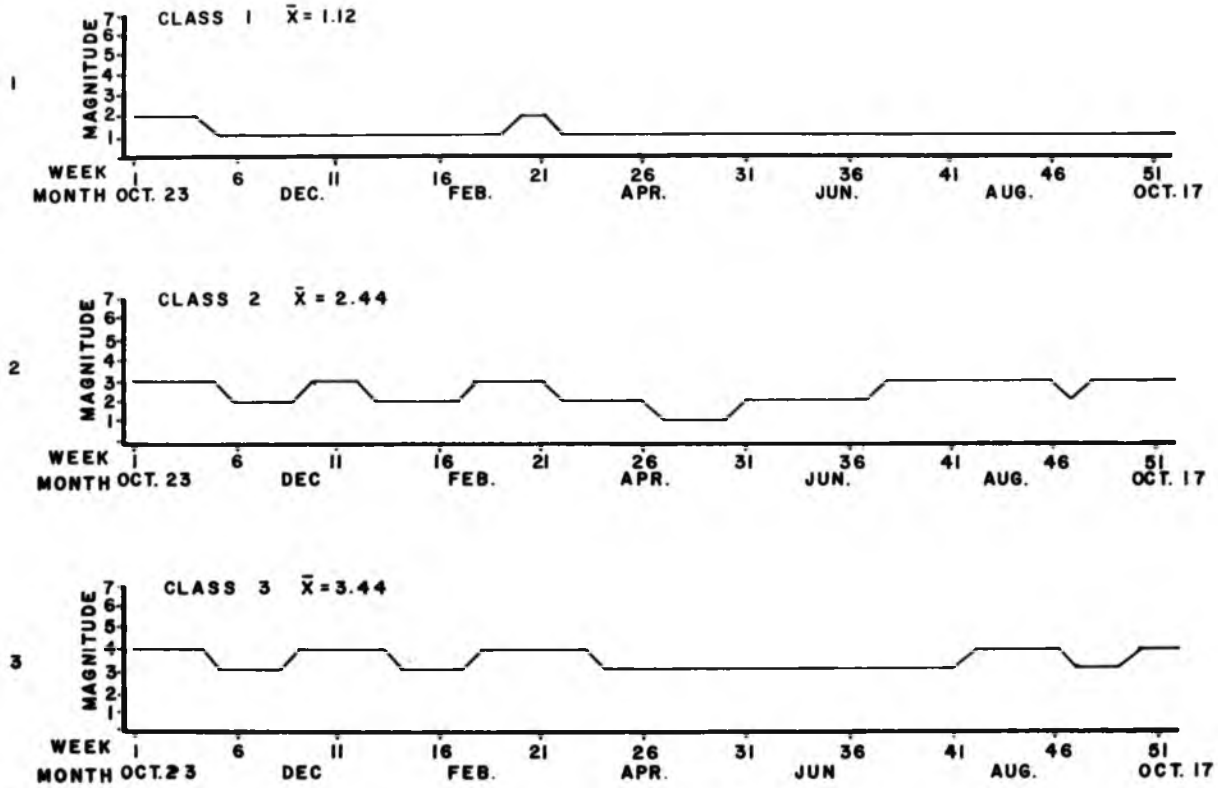
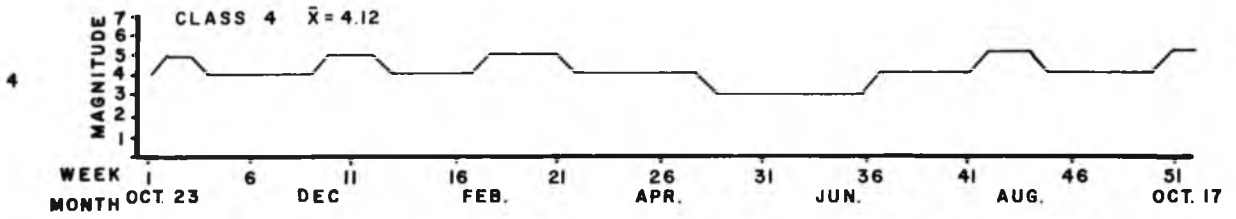

1
CLASS 1 $\bar{X}$ = 1.12
MAGNITUDE
7
6
5
4
3
2
1
WEEK
1
6
11
16
21
26
31
36
41
46
51
MONTH OCT. 23
DEC.
FEB.
APR.
JUN.
AUG.
OCT. 17
2
CLASS 2 $\bar{X}$ = 2.44
MAGNITUDE
WEEK
MONTH OCT. 23
DEC
FEB.
APR.
JUN.
AUG.
OCT. 17
3
CLASS 3 $\bar{X}$ = 3.44
MAGNITUDE
WEEK
MONTH OCT. 23
DEC
FEB.
APR.
JUN
AUG.
OCT. 17

PLATE I. (Continued)

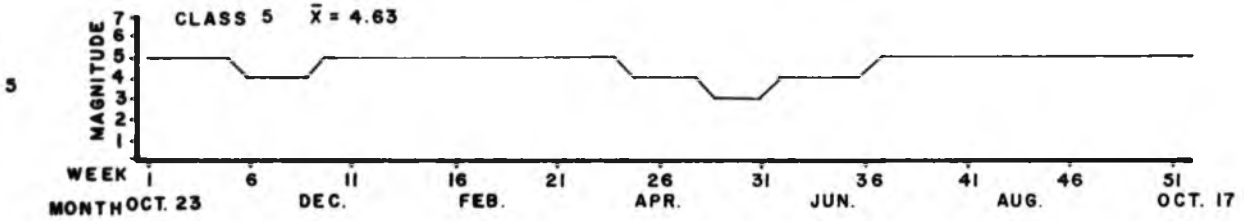
Figure 4. Class 4

Figure 5. Class 5

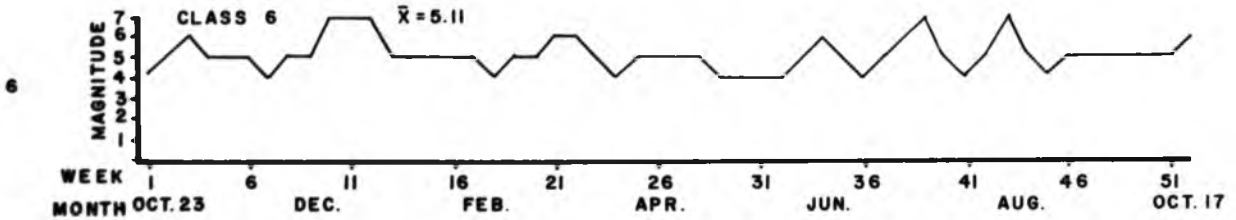
Figure 6. Class 6


CLASS 4 $\bar{X}$ = 4.12
MAGNITUDE
7
6
5
4
3
2
1
WEEK
1
6
11
16
21
26
31
36
41
46
51
MONTH
OCT. 23
DEC
FEB.
APR.
JUN.
AUG.
OCT. 17

4


CLASS 5 $\bar{X}$ = 4.63
MAGNITUDE
7
6
5
4
3
2
1
WEEK
1
6
11
16
21
26
31
36
41
46
51
MONTH
OCT. 23
DEC.
FEB.
APR.
JUN.
AUG.
OCT. 17

5


CLASS 6 $\bar{X}$ = 5.11
MAGNITUDE
7
6
5
4
3
2
1
WEEK
1
6
11
16
21
26
31
36
41
46
51
MONTH
OCT. 23
DEC.
FEB.
APR.
JUN.
AUG.
OCT. 17

6

The plants of Class 2 (Plate I, Fig. 2) were highly carpellodic but less so than those of Class 1 and showed a slightly greater variation in sexual expression. They varied in magnitude from 1 to 3 during the year and remained in Magnitude 1 (most carpellodic) for less than 20 per cent of the year.

The plants of Class 3 (Plate I, Fig. 3) were relatively stable in their sex expression, showing carpellody of Magnitudes 3 and 4 only. They never reached complete carpellody (Magnitude 1) nor produced an elongata flower (Magnitude 5). Slightly more than one half of the year was spent in Magnitude 3 and the rest in Magnitude 4.

Plants in Class 4 (Plate I, Fig. 4) varied from Magnitude 5 (normal elongata flowers) to Magnitude 3 (Pentandria). These plants produced normal elongata flowers for nearly one half of the year and slightly carpellodic flowers (Magnitude 4) and Pentandria flowers (Magnitude 3) for the rest of the year. Only one period during the year produced Magnitude 3 flowers (May-June).

The plants of Class 5 (Plate I, Fig. 5) were only very slightly carpellodic, remaining in Magnitude 5 (normal elongata flowers) for more than 70 per cent of the year. Magnitude 4 flowers were produced during three periods and Magnitude 3 flowers during May for one short period of only 3 weeks.

The plants of Class 6 (Plate I, Fig. 6) were those which showed slight shifts to either carpellody or sterility. The shifts were never longer than 3 weeks duration at a time and no more than a total of 12 weeks for the year in each direction. The shifts toward sterility were more severe than those toward carpellody (5 down to 4) in terms of the

number of magnitudes traversed, and in most cases reached the complete female sterility of Magnitude 7.

Class 7 (Plate I, Fig. 7) was made up of very slightly female sterile plants which showed normal elongata flowers for more than 70 per cent of the year and sterile flowers ranging from near normal (Magnitude 6) to completely sterile (Magnitude 7) for the remainder of the year.

Class 8 (Plate I, Fig. 8) was made up of plants producing normal flowers (Magnitude 5) for only about one half of the year and female sterile flowers from Magnitude 6 to Magnitude 7 for the rest.

The plants of Class 9 (Plate I, Fig. 9) produced normal flowers for less than 25 per cent of the year and shifted to complete female sterility (Magnitude 7) or lesser degrees of sterility (Magnitude 6) during the rest of the year.

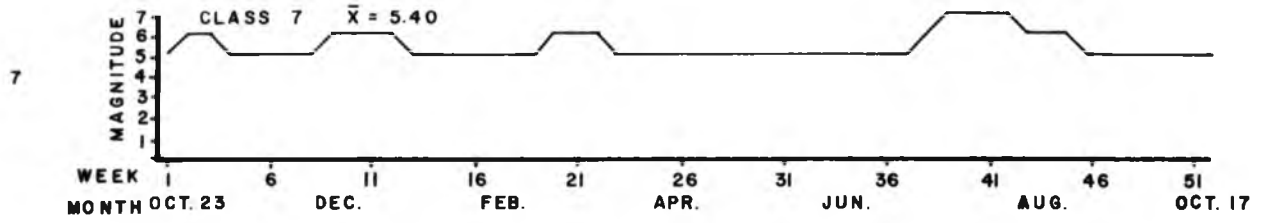
The plants of Class 10 (Plate I, Fig. 10) were highly sterile and produced a normal flower (Magnitude 5) only rarely. They remained completely female sterile (Magnitude 7) for nearly 70 per cent of the cycle and shifted to Magnitude 6 or 5 only during periods following lowest temperatures. These plants were highly stable but shifted to lesser degrees of sterility at times.

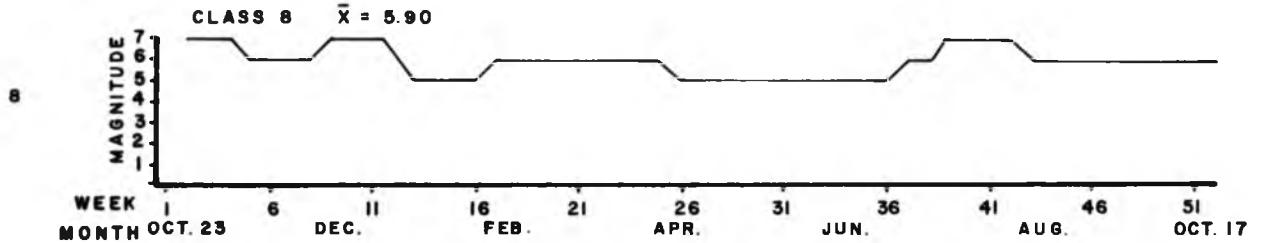
PLATE I. (Continued)

Figure 7. Class 7

Figure 8. Class 8

Figure 9. Class 9


7
CLASS 7 $\bar{X}$ = 5.40
MAGNITUDE
7
6
5
4
3
2
1
WEEK 1 6 11 16 21 26 31 36 41 46 51
MONTH OCT. 23 DEC. FEB. APR. JUN. AUG. OCT. 17


8
CLASS 8 $\bar{X}$ = 5.90
MAGNITUDE
7
6
5
4
3
2
1
WEEK 1 6 11 16 21 26 31 36 41 46 51
MONTH OCT. 23 DEC. FEB. APR. JUN. AUG. OCT. 17

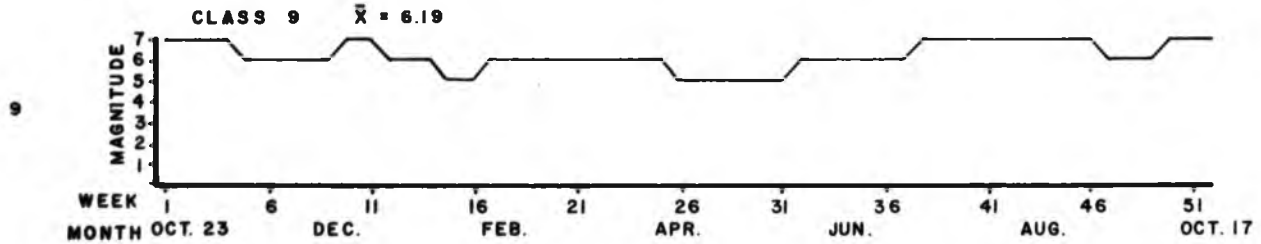
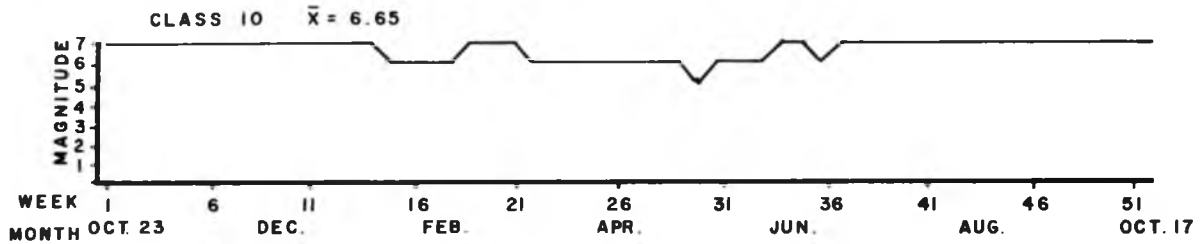

9
CLASS 9 $\bar{X}$ = 6.19
MAGNITUDE
7
6
5
4
3
2
1
WEEK 1 6 11 16 21 26 31 36 41 46 51
MONTH OCT. 23 DEC. FEB. APR. JUN. AUG. OCT. 17

PLATE I. (Continued)

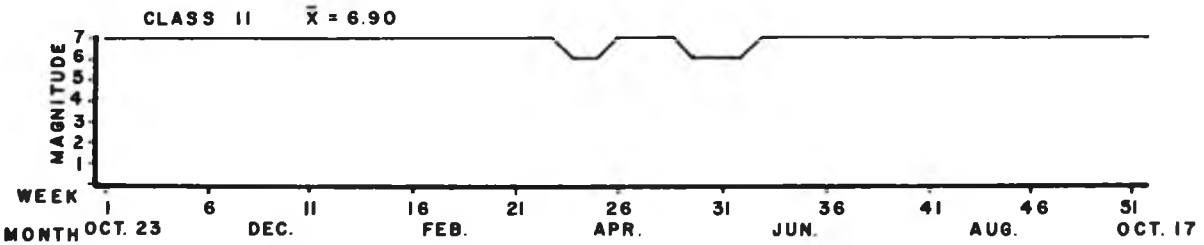
Figure 10. Class 10

Figure 11. Class 11

10


CLASS 10 $\bar{X}$ = 6.65
MAGNITUDE
7
6
5
4
3
2
1
WEEK
1
6
11
16
21
26
31
36
41
46
51
MONTH
OCT. 23
DEC.
FEB.
APR.
JUN.
AUG.
OCT. 17

11


CLASS 11 $\bar{X}$ = 6.90
MAGNITUDE
7
6
5
4
3
2
1
WEEK
1
6
11
16
21
26
31
36
41
46
51
MONTH
OCT. 23
DEC.
FEB.
APR.
JUN.
AUG.
OCT. 17

The plants of Class 11 (Plate I, Fig. 11) were the most sterile of all plants in the orchard. They remained in Magnitude 7 (completely sterile) for more than 90 per cent of the year and when shifts occurred, they were of short duration and of only one magnitude toward lesser sterility. These plants were extremely stable in sex expression and a large proportion of them were from parental Line 13BF6.

Histograms representing sex variation in the plants found in the 11 classes show the differences between classes from the maximum levels of carpellody (Class 1) to sterility (Class 11) and the duration of time the plants spent in each magnitude (Plate II, Figs. 12 - 22).

D. FLORAL INITIATION AND MORPHOLOGY

The time between floral initiation and anthesis in papaya was unknown at the time this experiment was started. Therefore, it became necessary to determine when the flower began its ontogeny: i.e., the interval between inception and anthesis. This information would provide more accurate correlation between climatological data and sex expression. In Carica papaya L. one flower panicle is produced in the axil of each leaf and leaves are produced in a regular phylotaxy (3). First, the rate of flower production was determined. This was done by noting the number of new flowers opened over a period of time, in this case 5 weeks during the fall season from October 22 to November 19 and the number of unopened buds remaining on the plant from the apex down to the last opened flower (Table IV). The apex was taken to be the smallest emerged bud visible at the very top of the plant and required the use of a small magnifying

PLATE II

Variability in magnitude and number of weeks in each magnitude of classes 1 through 11

Figure 12. Class 1

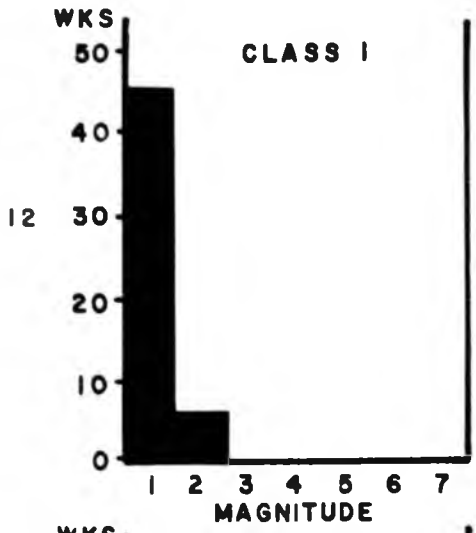
Figure 13. Class 2

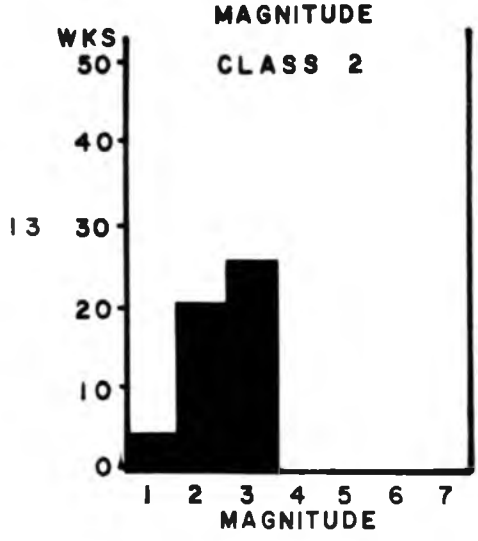
Figure 14. Class 3

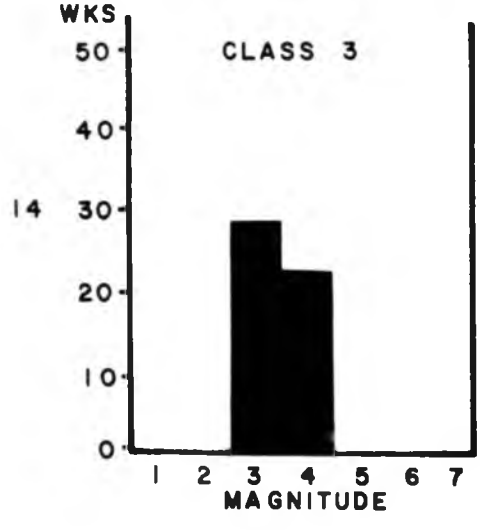
Figure 15. Class 4

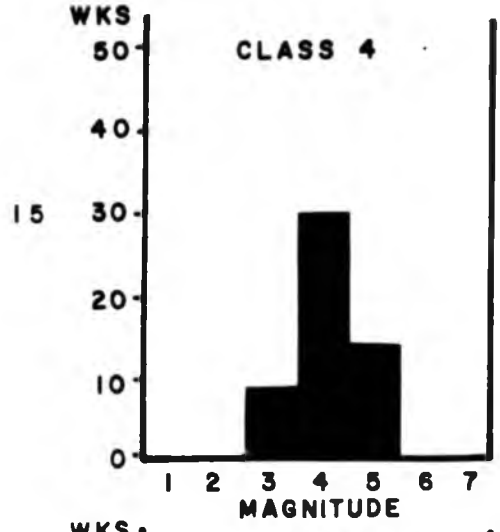
Figure 16. Class 5

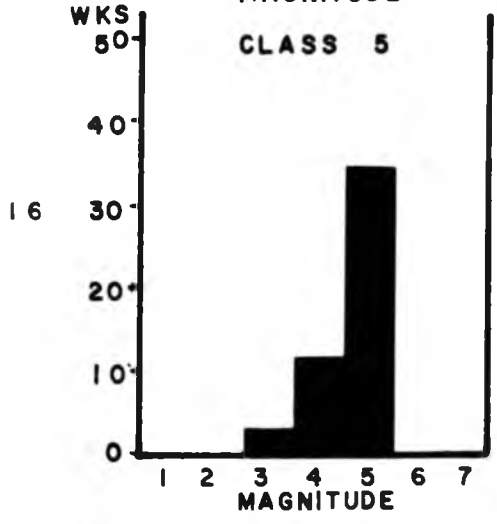
Figure 17. Class 6


WKS
CLASS 1
50
40
30
20
10
0
12
1 2 3 4 5 6 7
MAGNITUDE


WKS
CLASS 2
50
40
30
20
10
0
13
1 2 3 4 5 6 7
MAGNITUDE


WKS
CLASS 3
50
40
30
20
10
0
14
1 2 3 4 5 6 7
MAGNITUDE


WKS
CLASS 4
50
40
30
20
10
0
15
1 2 3 4 5 6 7
MAGNITUDE


WKS
CLASS 5
50
40
30
20
10
0
16
1 2 3 4 5 6 7
MAGNITUDE

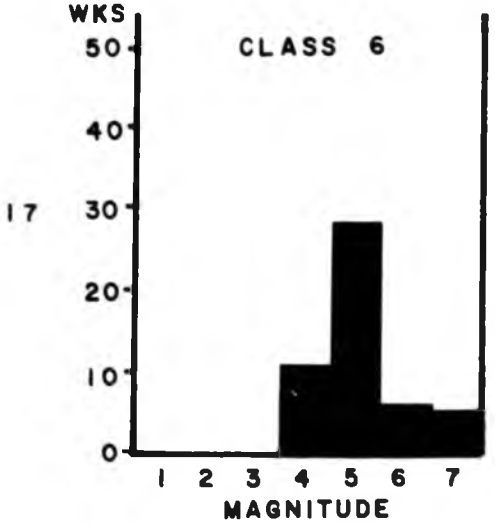

WKS
CLASS 6
50
40
30
20
10
0
17
1 2 3 4 5 6 7
MAGNITUDE

PLATE II. (Continued)

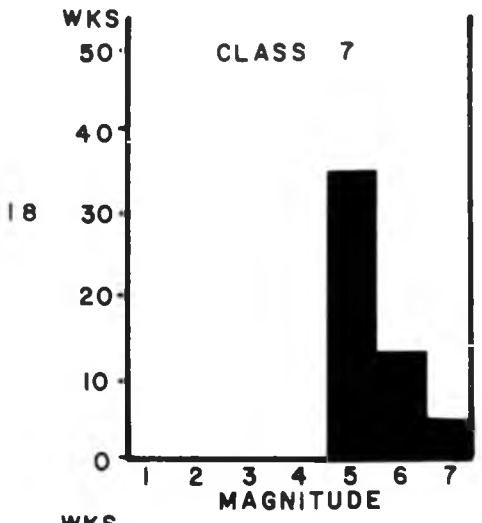
Figure 18. Class 7

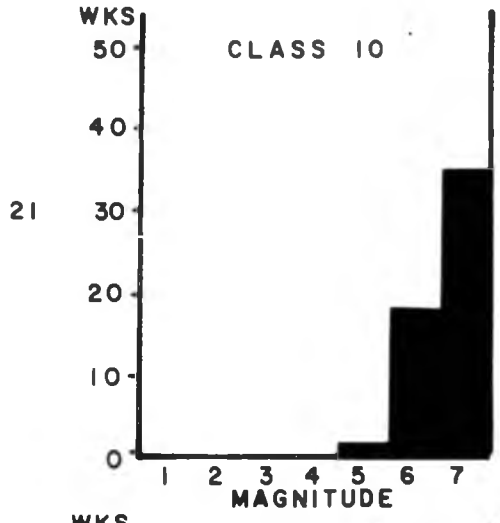
Figure 19. Class 8

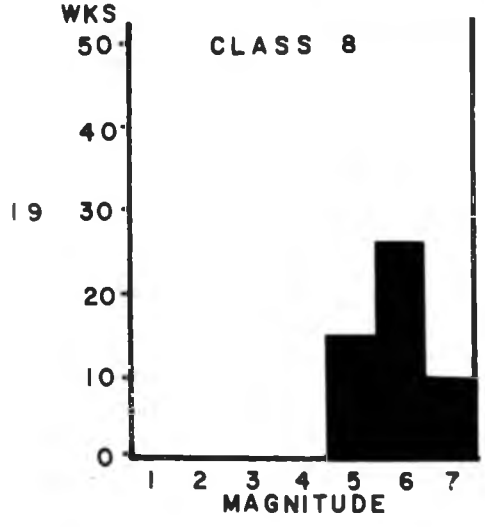
Figure 20. Class 9

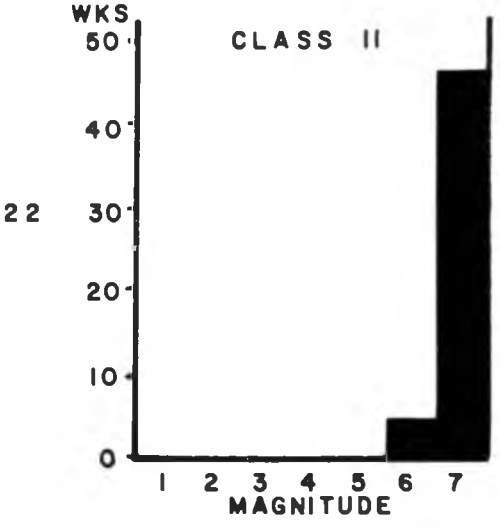
Figure 21. Class 10

Figure 22. Class 11


WKS
CLASS 7
50
40
18 30
20
10
0
1 2 3 4 5 6 7
MAGNITUDE


WKS
CLASS 10
50
40
21 30
20
10
0
1 2 3 4 5 6 7
MAGNITUDE


WKS
CLASS 8
50
40
19 30
20
10
0
1 2 3 4 5 6 7
MAGNITUDE


WKS
CLASS 11
50
40
22 30
20
10
0
1 2 3 4 5 6 7
MAGNITUDE

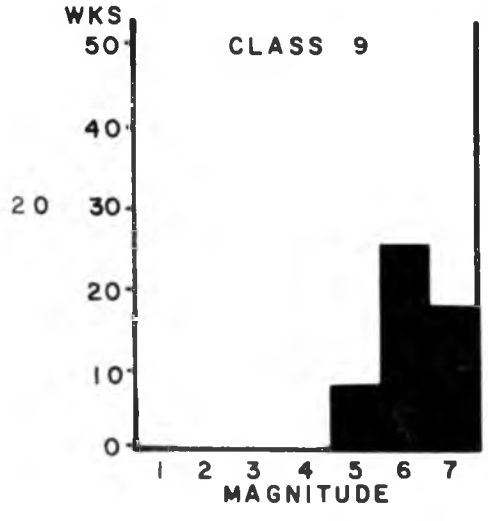

WKS
CLASS 9
50
40
20 30
20
10
0
1 2 3 4 5 6 7
MAGNITUDE

TABLE IV. THE NUMBER OF FLOWERS REACHING ANTHESIS EACH WEEK OVER (/) THE NUMBER OF UNOPENED BUDS

PLANT NUMBER AND TYPE	DATE OCT 22	OCT 29	NOV 5	NOV 12	NOV 19	TOTAL NEW FLOWERS/ AVERAGE NUMBER OF UNOPENED BUDS
1 normal	4/20	3/22	2/21	2/22	2/22	13/21.4
2 "	2/21	1/20	2/21	3/21	2/20	10/20.6
3 "	3/22	1/20	2/20	4/21	1/21	11/20.8
4 "	1/20	2/20	4/21	3/22	3/23	13/21.2
5 "	4/21	2/21	2/21	1/20	2/21	11/20.8
6 "	2/19	2/19	3/20	2/21	2/22	11/20.2
7 "	1/20	4/19	1/20	3/21	1/22	10/20.4
8 "	3/20	2/20	1/20	3/22	3/21	12/20.6
9 "	2/20	4/21	0/21	3/21	2/21	11/20.8
10 "	3/23	2/22	2/21	4/23	1/23	12/22.4
11 "	1/21	4/20	2/21	4/22	2/23	13/21.4
12 "	1/21	4/21	2/21	3/21	4/22	14/21.2
13 "	1/18	3/19	2/19	3/20	2/20	11/19.2
14 "	1/19	2/19	2/20	4/21	2/21	11/20.0
15 "	1/19	3/19	3/21	2/20	2/21	11/20.0
16 normal	2/20	3/20	2/20	4/21	1/21	12/20.4
17 carpellodic	1/19	4/20	1/20	4/22	1/22	11/20.6
18 carpellodic	2/21	4/20	2/21	2/23	3/23	13/21.6
19 sterile	3/21	2/21	2/21	2/21	3/22	12/21.2
20 "	4/22	2/23	3/23	4/24	2/22	15/22.8
21 "	2/21	3/20	3/21	2/22	2/22	12/21.2
22 "	1/17	3/18	2/18	0/18	5/20	11/18.2
23 "	2/19	2/19	2/19	3/21	2/21	11/19.8
24 sterile	2/20	3/21	3/21	2/21	3/22	13/21.0
25 female	3/20	3/21	1/21	4/21	1/22	12/21.0
26 "	3/21	2/21	3/21	3/21	3/22	14/21.2
27 "	2/20	3/21	2/22	3/22	3/22	13/21.4
28 female	2/22	6/22	3/24	1/22	1/22	13/22.4
29 male	3/16	3/16	0/16	2/18	1/19	9/17.0
30 male	4/17	6/20	2/21	3/23	1/22	16/20.6

glass to observe. An average was obtained for the number of flowers to reach anthesis during the 5 week period on the five different types of plants and was found to range from 11.81 on the normal types to 12.75 on the female types (Table V). Analysis of variance showed this difference to be non-significant ($F = 0.29$). Although the rate of flower production may be different at a different season of the year, it is unlikely that the plants tested would respond differently.

A range for the mean number of flowers opened during the five weeks was obtained by adding or subtracting the standard deviations of each mean (Table V). The minimum, mean, and maximum figures for the number of new flowers opened were then divided into 35, the number of days in the test period and the resultant figures yielded a maximum, mean, and minimum number of days required to produce one new flower (Table VI). These figures were then multiplied by the number of unopened flower buds (Table VI) and yielded a range (minimum, mean, and maximum) for the number of days required from initiation to anthesis. These figures indicate that the plants in the test required from 7 to 10 weeks to reach anthesis from their inception. The 3 hermaphroditic lines tested (normal, carpellodic, and sterile) showed no significant variation in the length of time required from floral inception to anthesis.

In addition to determining the approximate time of floral initiation, morphological features of the various sex types of plants used in the experiments were compared. This was done to give some indication of the time of occurrence of sexual changes and the degree of change occurring during the period of ontogeny of the flowers. Flowers for this study were gathered from the plots at Malama-Ki. Histological sections were

TABLE V. FLOWERS REACHING ANTHESIS DURING A FIVE WEEK PERIOD FROM OCTOBER 22 TO NOVEMBER 19

PLANT TYPE	TOTAL NO. OF PLANTS	TOTAL NO. OF FLOWERS	MEAN NO. FLOWERS REACHING ANTHESIS ±S.D.[a]
HERMAPHRODITIC			
NORMAL	16	189	11.81 ± 1.09
CARPELLODIC	2	24	12.00 ± 1.41
STERILE	6	72	12.00 ± 2.00
DIOECIOUS			
FEMALE	4	51	12.75 ± 1.21
MALE	2	25	12.50 ± 4.91

[a]Not significant

TABLE VI. NUMBER OF UNOPENED FLOWERS, TIME REQUIRED TO PRODUCE ONE NEW FLOWER AND AVERAGE NUMBER OF DAYS REQUIRED FROM FLORAL INITIATION TO ANTHESIS

SEX TYPE	NO. UNOPENED FLOWERS	DAYS REQUIRED TO PRODUCE ONE NEW FLOWER			NO. OF DAYS FROM INITIATION TO ANTHESIS		
		MIN.	MEAN	MAX.	MIN.	MEAN	MAX.
HERMAPHRODITIC							
NORMAL	20.68	2.71	2.99	3.26	56	61	67
CARPELLODIC	21.10	2.61	2.96	3.30	55	62	70
STERILE	20.73	2.50	3.00	3.50	52	61	73
DIOECIOUS							
FEMALE	21.43	2.51	2.77	3.03	54	59	65
MALE	18.76	2.01	3.31	4.61	38	53	86

made of the meristematic regions of all tree types and of flowers of all types from initial stage to maturity. Tissues to be examined were selected from actively growing plants of the desired sex type and immediately killed and fixed in FAA (formalin-aceto-alcohol). They were then dehydrated in a tertiary butyl alcohol series as described by Johansen (52) and Sass (85). Tissues were embedded in paraffin and tissuemat and sectioned at a thickness of 10 to 15 microns on a rotary microtome. All sections were stained with safranin and fast green and permanently mounted on glass micro slides (52, 85). Photomicrographs were prepared to illustrate this portion of the study. Histological studies of the developmental sequence of the various flower parts and the differences between the different sex types were undertaken to determine the time when the floral parts are most susceptible to environmental influences.

E. CORRELATING SEX CLASSES WITH ENVIRONMENTAL FACTORS

It has already been shown by other workers (5, 6, 7, 8, 62, 63) that temperature is the main influence on sex changes and that other factors such as humidity, sunlight, photoperiod, nutrition, and physical damage all had a lesser effect on sex changes. Previous studies also showed (22, 70, 88) that moisture levels affected sex expression rather strongly.

For this reason, temperature and rainfall data were plotted on a weekly basis for correlation with sex changes occurring in the plants. The weekly mean minimum and mean maximum temperatures; the difference between the mean minimum and maximum temperatures; and the total weekly

rainfall are charted in Plate III. The "0" point on the horizontal axis of each graph indicates the point at which the first flowers reached anthesis and taking of sex expression data began. Data for temperature and rainfall were collected from 10 weeks prior to when initial flower data were recorded to one year after. It was suspected that temperature may be the primary influencing factor on sex expression, and, further, that this influence occurred during the flower bud initiation and early developmental stages. Climatic data were recorded for 10 weeks prior to anthesis to insure that this data would be available during the initiation stage of the first flowers, which at this point in the experiment was unknown.

The performance of each class of plants (Classes 1 through 11, Plate I) was correlated to each of the temperature and rainfall graphs (Plate III). Previous anatomical studies indicated that sexual changes in the flowers occurred 4 to 10 weeks prior to anthesis. By 4 weeks before anthesis, the flower parts were in their final form. Correlation of each class with a temperature or rainfall graph was begun by placing the week 1 position on a class graph over the -4 week position on a temperature or rainfall graph. The fit (correlation) of the two graphs was then calculated for winter conditions during October through March and for summer conditions during April through September. The week 1 position on the class graph was then shifted back along the temperature or rainfall graph to the -5 week position and correlations calculated again. This process was repeated for each class graph and each climate graph from 4 to 10 weeks prior to anthesis. The week at which the best correlation was obtained for each pair of graphs correlated would give

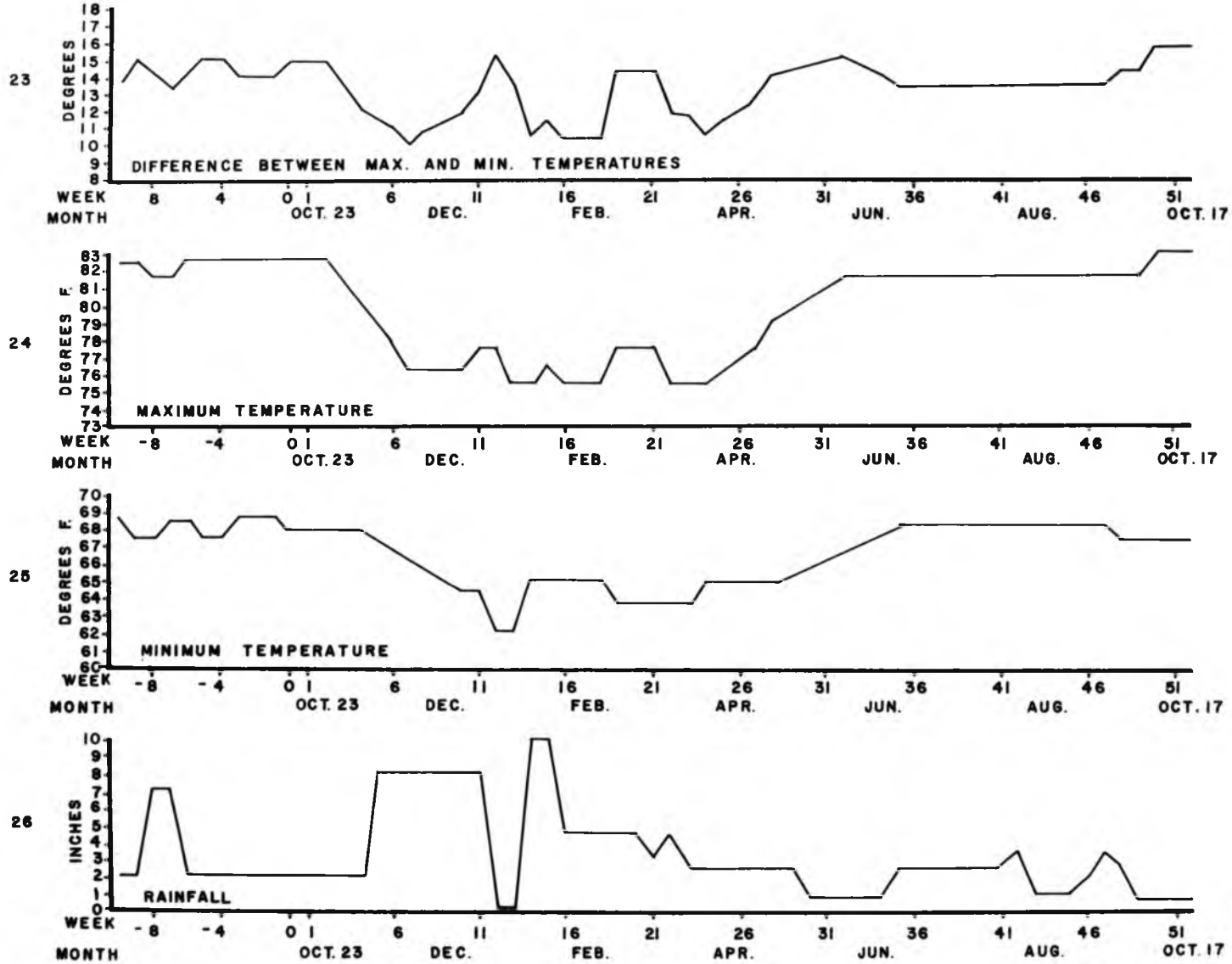
PLATE III

Figure 23. Difference between maximum and minimum temperature (mean weekly) for experimental period at Malama-Ki substation

Figure 24. Maximum temperature (mean weekly) for experimental period at Malama-Ki substation

Figure 25. Minimum temperature (mean weekly) for experimental period at Malama-Ki substation

Figure 26. Rainfall pattern (total weekly amount) for experimental period at Malama-Ki substation


23
DEGREES
DIFFERENCE BETWEEN MAX. AND MIN. TEMPERATURES
24
DEGREES F.
MAXIMUM TEMPERATURE
25
DEGREES F.
MINIMUM TEMPERATURE
26
INCHES
RAINFALL
WEEK
MONTH
OCT. 23
DEC.
FEB.
APR.
JUN.
AUG.
OCT. 17

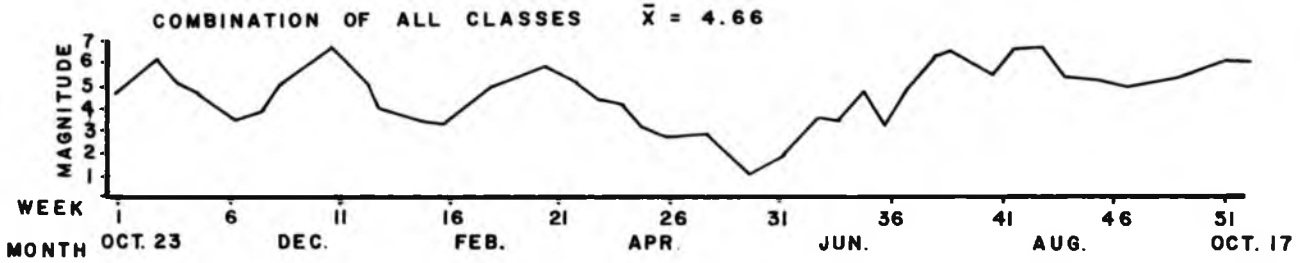
an indication that this was the point at which the sexual organs of the plant were most susceptible to changes influenced by temperature and rainfall (Table VII).

One other correlation was conducted. All classes of plants go through approximately the same general pattern of sex change during the year. When temperatures decline, the magnitude of sex expression of all plants tends to decline also; carpellodic plants become more carpellodic and sterile plants become less sterile. When temperatures rise, carpellodic plants become less carpellodic and sterile plants become more sterile. Thus all plants of all progenies were combined into one single curve showing both carpellody and sterility (Plate IV, Fig. 27). The magnitudes of sex expression represented by the plants of classes 1 through 11 were added together for each week during the test period. This yielded a range of magnitudes from the lowest to the highest in the entire orchard for each week from week 1 to week 52. Total added magnitudes ranged from 51 to 85. During the coldest periods each plant would be at its lowest on the magnitude scale and during the warmer period, each plant would be at its highest on the scale. On this basis, then, a magnitude of 1 was assigned to the lowest figure of the added magnitude range, 51, and a magnitude of 7 was assigned to the highest figure of the added magnitude range, 85. All points between 51 and 85 were assigned magnitudes ranging from 1 to 7 in order to be able to plot the combined curve on the same scale as classes 1 through 11. This curve represents the average state of sexual expression of all the plants in the test plot over the year's cycle.

PLATE IV

Figure 27. Variability in magnitude of the entire combined orchard over a 52 week period

27


COMBINATION OF ALL CLASSES
$\bar{X}$ = 4.66
MAGNITUDE
7
6
5
4
3
2
1
WEEK
1
6
11
16
21
26
31
36
41
46
51
MONTH
OCT. 23
DEC.
FEB.
APR.
JUN.
AUG.
OCT. 17

The sexual response of each class (1 through 11) to temperature changes during the year was also plotted to determine the rate or speed of response and the amount of response in each class.

F. GENETIC STUDIES

Bar graphs were constructed to show the segregation patterns of carpellody and sterility in progenies of the inbred parental lines and were also used to determine the mode of inheritance of these characteristics by their progenies. A genetic analysis of the parental and progeny lines was made. Genetic analysis was carried out by comparing the population means of the progenies with the population means of their parents. Variance was also used (95) to indicate the mode of inheritance of the characters, carpellody and sterility.

RESULTS AND DISCUSSION

A. FLORAL INITIATION AND MORPHOLOGY

The progressive ontogeny of a typical hermaphroditic flower (Plate V) indicates that stamen and ovary differentiation apparently does not occur until after the flower bud becomes visible in a leaf axil. The first flower becomes visible as a bud in a leaf axil (tangential view) near the top of the plant at approximately leaf node number 3 (Plate V, Fig. 28). A longitudinal view of leaf node 2 or 3 on the left side of the photo (Plate V, Fig. 29) and node 4 on the right side shows that the floral bud is not fully visible to the naked eye prior to leaf node 4. At approximately leaf node 5 only sepals and the beginning corolla tube have differentiated (Plate V, Fig. 30). At about node 6 floral organs have not yet differentiated (Plate V, Fig. 31). At about leaf node 7 the first stamen initials are just becoming visible (Plate V, Fig. 32). At about node 8 stamens are slightly more developed and are beginning to show form (Plate V, Fig. 33). Another flower at leaf node 8 (Plate V (Con't.), Fig. 34) and of about the same age as the flower in Figure 33 is from a highly carpellodic plant. In this case, the developing stamens appear to be larger in width than those in Figure 33. Although the figure is too small for accurate determination, it appears that the supportive stamen structures may not be as "normally" developed as those in Figure 33. A normal flower at about leaf node 10 or 11 (Plate V (Con't.), Fig. 35) has stamens nearly completely developed and the first initials of the new ovary can be seen. A normal flower at about leaf node 13 or 14 (Plate V (Con't.), Fig. 36) has floral organs that are nearly

PLATE V

Figure 28. Tangential view of young flower bud just appearing in leaf axil number 3.

Figure 29. Longitudinal view of emerging flower buds in leaf axil number 2 or 3 on the left and number 4 on the right.

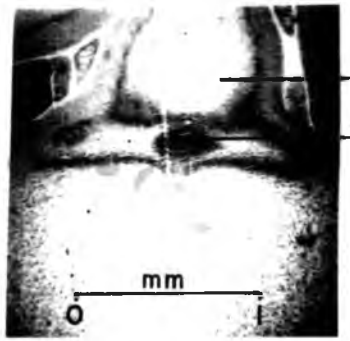
Figure 30. Longitudinal view of young flower bud at about leaf node 5.

Figure 31. Longitudinal view of young flower bud at about leaf node 6.

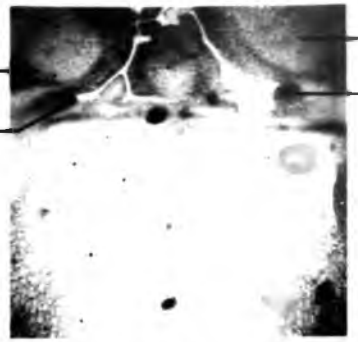
Figure 32. Longitudinal view of young flower bud at about leaf node 7.

Figure 33. Longitudinal view of young flower bud at about leaf node 8.

Lp - leaf primordia
Eb - emerging flower bud
Se - sepals
Ct - corolla tube
Es - emerging stamens



28



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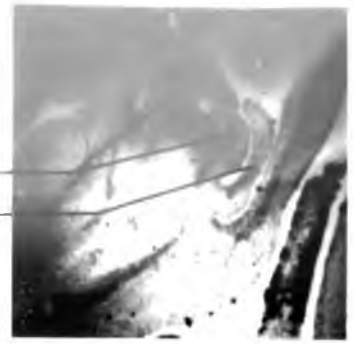

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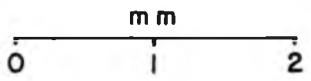
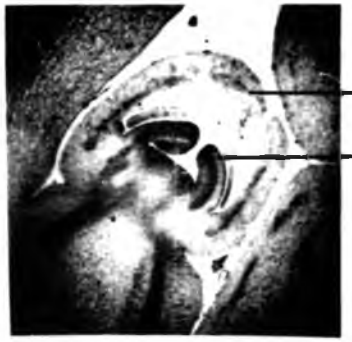
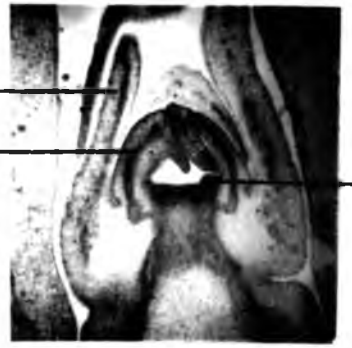




31



32

33

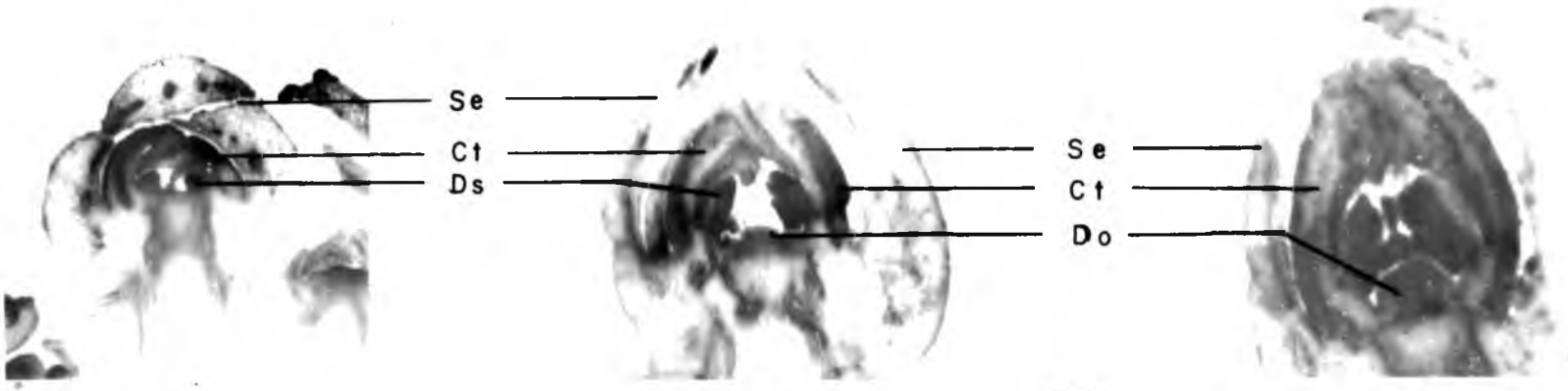
PLATE V (Continued)

Figure 34. Longitudinal view of young flower bud from approximately leaf node 8 from a highly carpellodic plant.

Figure 35. Longitudinal view of a young flower bud at about leaf node 10 or 11 from a normal hermaphroditic plant.

Figure 36. Longitudinal view of a young flower bud at about leaf node 13 or 14 from a normal hermaphroditic plant.

Se - sepals
Ct - corolla tube
Do - developing ovary
Ds - developing stamens


Se
Ct
Ds
Se
Ct
Do
34
35
mm
0
1
2
36

completely developed. It appears that flowers may require about 5 weeks to reach the stage indicated in Figure 36.

Stamens appear to begin differentiation shortly after the flower bud becomes visible, and have reached their final shape by the third week (node 10 or 11) (Plate V (Con't.), Fig. 35). At this point the ovary is just becoming a discernable feature, having appeared as a small mass of cells during the second week (node 8).

Microscopic examination of flowers during all stages of growth from initiation to anthesis showed that the floral structures are completely differentiated by the fifth week. Five week old male, female, and hermaphroditic flowers (Plates VI, VII, and VIII) clearly show the various structures in their completed forms. In the male and female flowers (Plates VI and VII) the organs shown are never transposed or altered in any way. Their ontogeny is apparently complete in 5 to 6 weeks. The hermaphroditic flower (Plate VIII) is also apparently complete in about 5 weeks. In the case of this flower, however, the ovary and stamens may be altered into the carpellodic or female sterile states at an early stage of development (Plates IX and X).

Five week old hermaphroditic flowers which have become female sterile and moderately carpellodic, respectively (Plates IX and X) indicate the similarity of features between these two types and the true male and female types (Plates VI and VII). The true male (Plate VI) and the hermaphroditic female sterile flower (Plate IX) show similarity in their gross appearance although subtle differences in petal and tubular texture may be noted. The vestigial ovaries of each type are somewhat different in size, that of the true male being more elongated

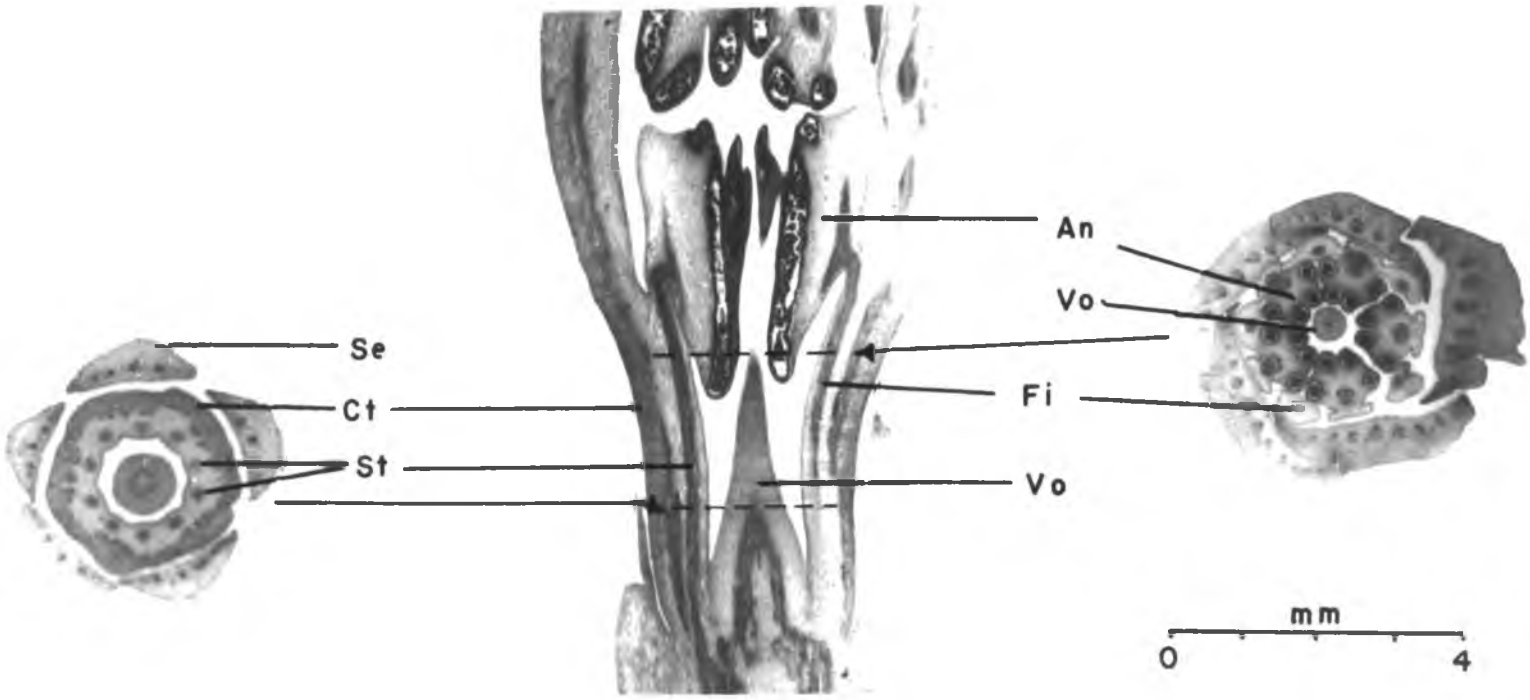
PLATE VI

Figure 37. Cross section near the base of a typical male flower in its fifth week of development.

Figure 38. Median longitudinal section of a typical male flower in its fifth week of development.

Figure 39. Cross section of the flower shown in Figure 37 but further up the corolla tube as indicated.

An - anther
Fi - stamen filament
Vo - vestigial ovary
Se - sepals
Ct - corolla tube
St - stamen traces



37 38 39

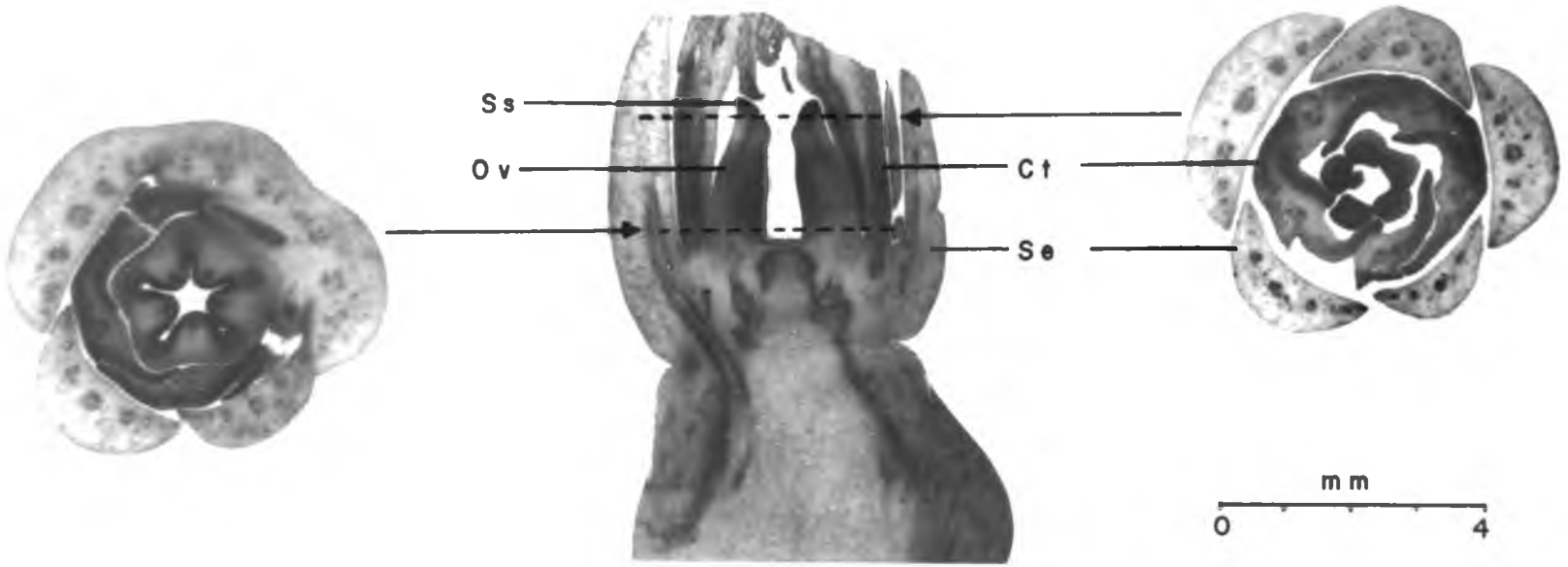
PLATE VII

Figure 40. Cross section near the base of a typical female flower in its fifth week of development.

Figure 41. Median longitudinal section of a typical female flower in its fifth week of development.

Figure 42. Cross section of a typical female flower as shown in Figure 40 but further up the corolla tube as indicated.

Ov - Ovary
Ct - corolla tube
Se - sepals
Ss - stigmatic surface



40 41 42

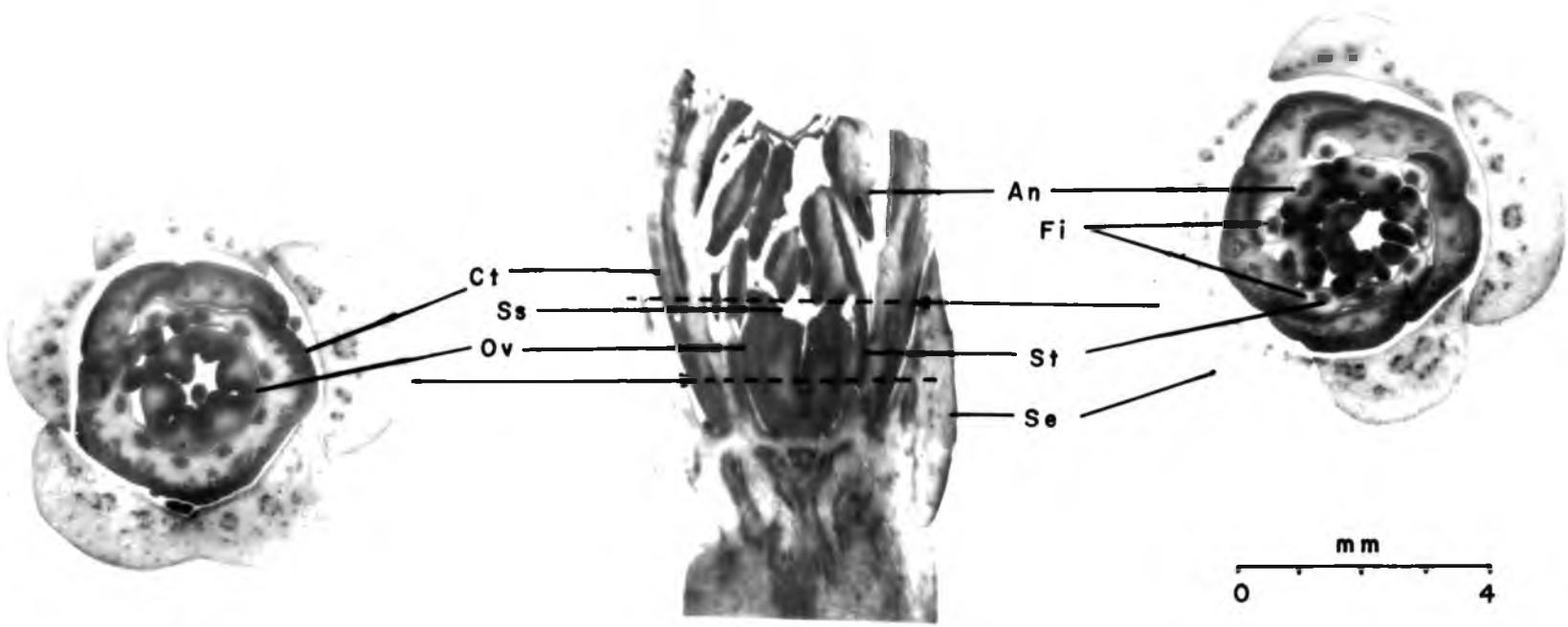
PLATE VIII

Figure 43. Cross section near the base of a normal hermaphroditic flower in its fifth week of development.

Figure 44. Median longitudinal section of a normal hermaphroditic flower in its fifth week of development.

Figure 45. Cross section of a normal hermaphroditic flower as shown in Figure 43 but further up the corolla tube.

An - anther
Ss - stimatic surface
Ov - ovary
Ct - corolla tube
Se - sepals
Fi - filaments
St - stamen traces



43 44 45

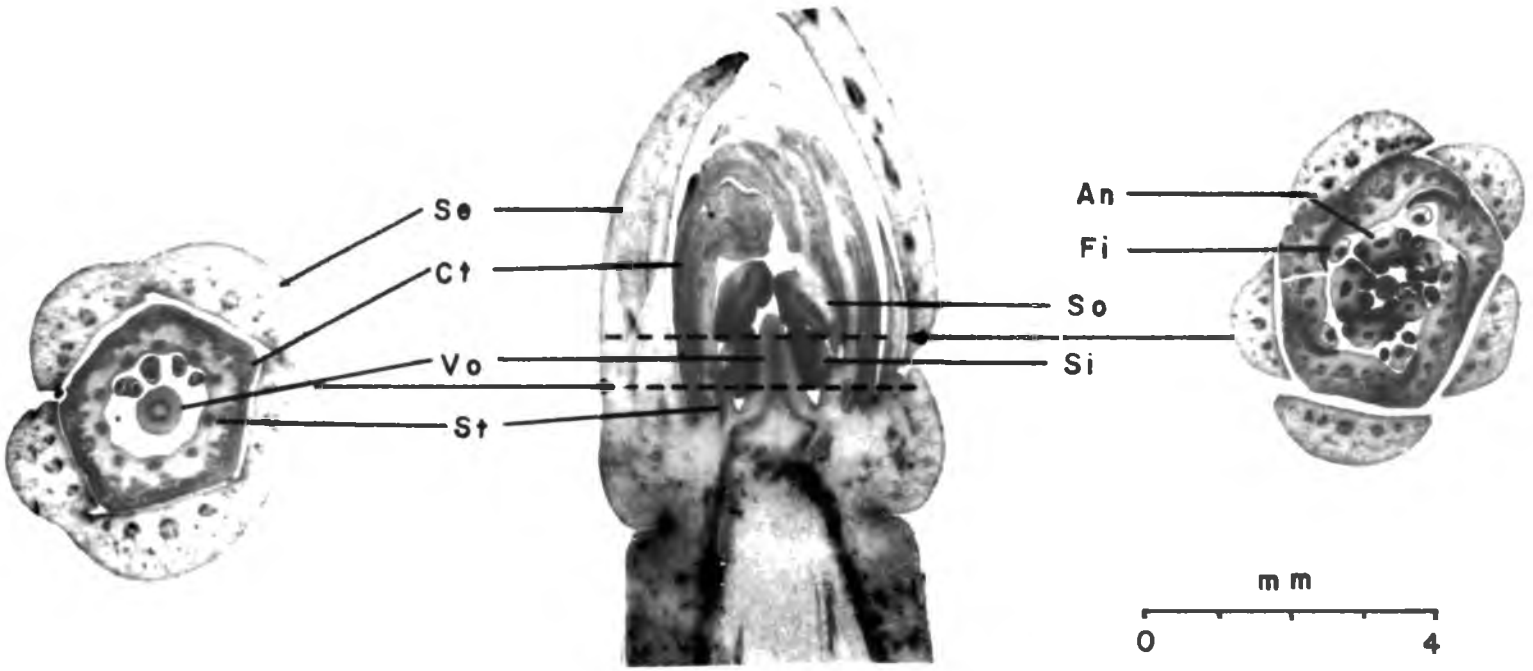
PLATE IX

Figure 46. Cross section near the base of a female sterile hermaphroditic flower in its fifth week of development.

Figure 47. Median longitudinal section of a female sterile hermaphroditic flower in its fifth week of development.

Figure 48. Cross section of female sterile hermaphroditic flower as shown in Figure 46 but further up the corolla tube.

Se - sepals
Ct - corolla tube
Vo - vestigial ovary
St - stamen trace
Fi - filament
So - stamen of outer whorl
An - anther
Si - stamen of inner whorl



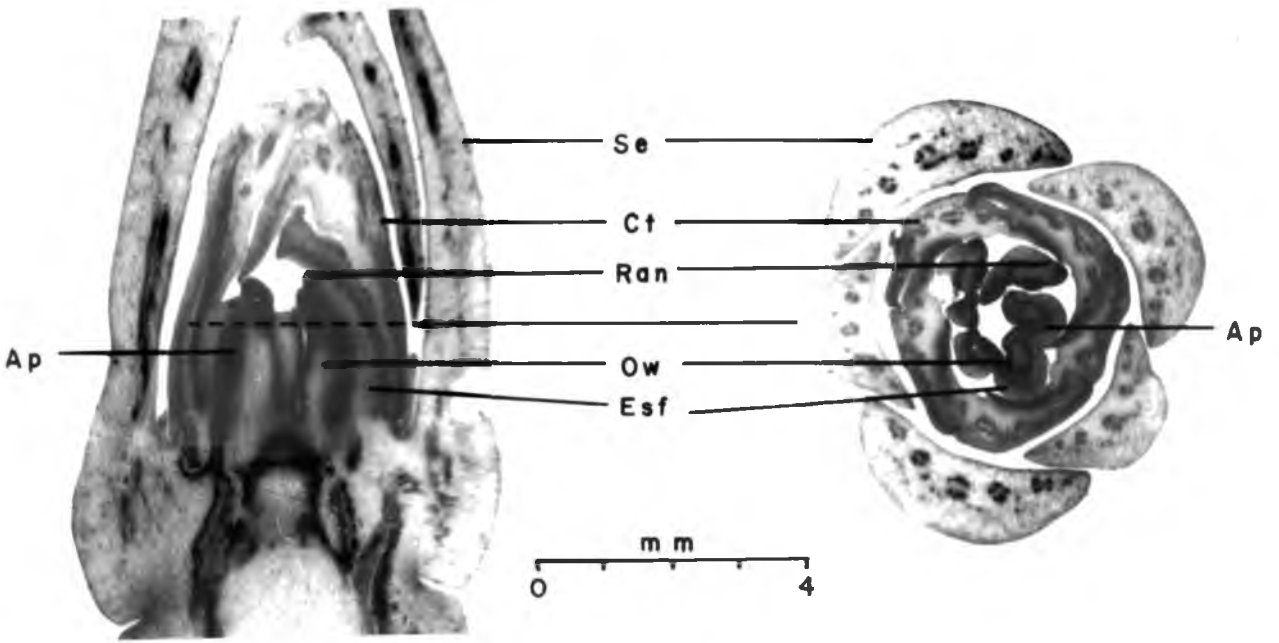
46 47 48

PLATE X

Figure 49. Median longitudinal section of a moderately carpellodic flower in its fifth week of development.

Figure 50. Median cross section of a moderately carpellodic flower in its fifth week of development.

Ran - reduced anther head
Se - sepals
Ct - corolla tube
Ow - ovary wall (original)
Ap - adnation point
Esf - expanded stamen filaments



49

50

than that of the sterile hermaphrodite. The sexual parts of the true male and sterile hermaphrodite flowers reach approximately the same degree of differentiation and the same general form at about the same time. The same ontogenetic sequence seems to occur in both types of flower.

A hermaphroditic flower in a moderately carpellodic state (Plate X) shows the basic ovarian structure. The stamen filaments, however, have become highly expanded and appear to be adnate to the ovary. The anther head on the right side of the flower (Plate X, Fig. 49) is no longer discernable while a small knob representing arrested development of the anther head on the left side is still visible. The stimulus to develop a normal anther head was either suppressed or absent.

This study shows the ovary to begin its ontogeny about 2 weeks after the stamens begin their ontogeny (Plate V). The suggested relative sequence of events in the development of a flower from its inception to anthesis is summarized in Figure 51 (Plate XI).

Therefore, since ovary differentiation starts about 2 weeks later than stamen differentiation, any changes caused by temperature would be expected to be apparent sooner in the stamens than in the ovaries.

B. EFFECTS OF TEMPERATURES ON INBRED LINES

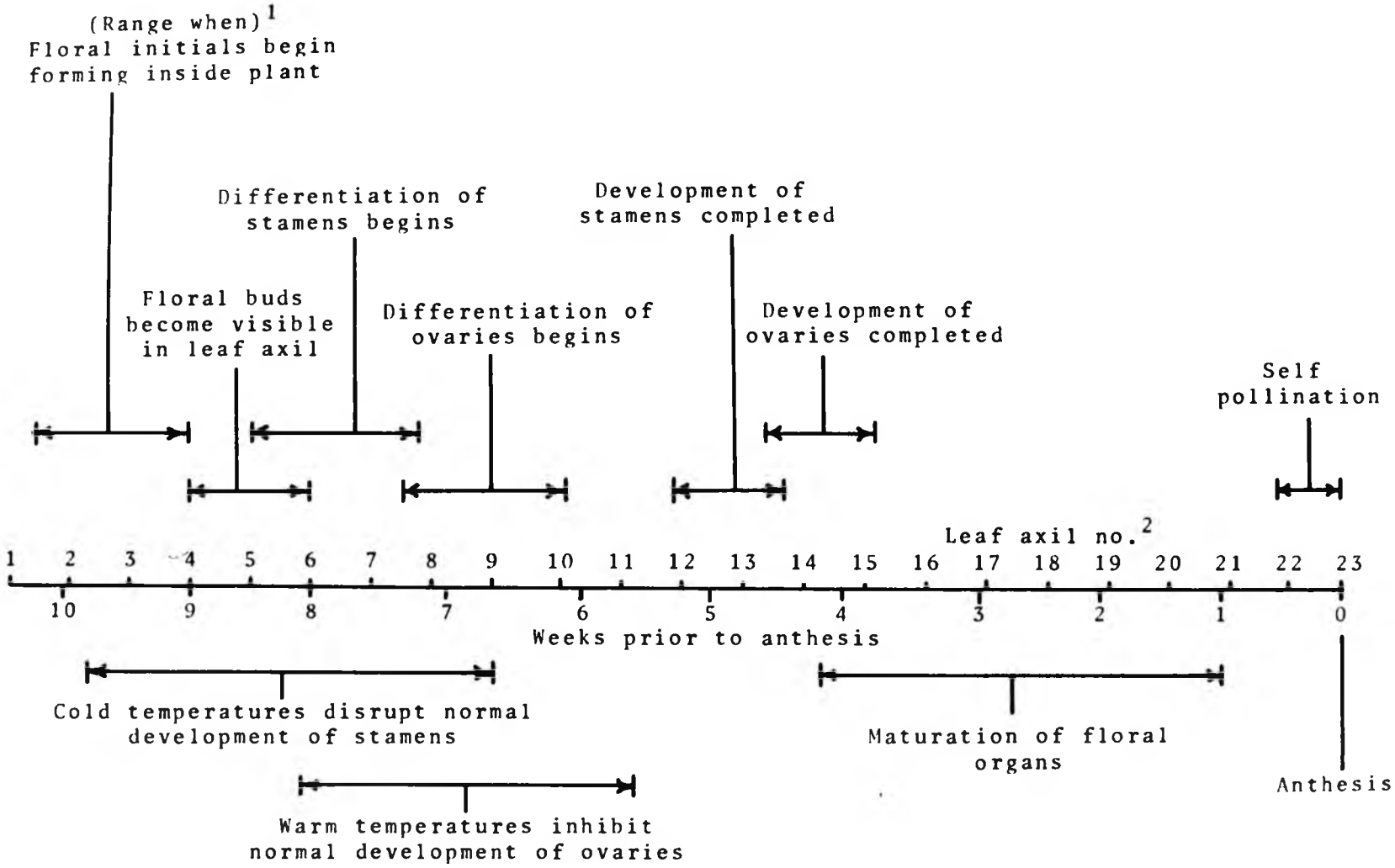
The 3 inbred highly stable lines (17CF6 - highly carpellodic, 17AF6 - low carpellody/low sterile (normal), and 13BF6 - highly sterile) were subjected to high, low, and normal temperatures to measure the response of known phenotypes to different temperatures. The experiment was started in January and continued for 30 weeks beyond which time the

PLATE XI

Figure 51. Suggested time sequence of developmental stages of floral organs prior to anthesis.

[1]All processes are given as ranges of time.

[2]Leaf axil number is given to indicate the nodal position on the average plant, from apical meristem down to the first open flower.


(Range when)[1]
Floral initials begin forming inside plant
Differentiation of stamens begins
Development of stamens completed
Floral buds become visible in leaf axil
Differentiation of ovaries begins
Development of ovaries completed
Self pollination
Leaf axil no.[2]
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23
10 9 8 7 6 5 4 3 2 1 0
Weeks prior to anthesis
Cold temperatures disrupt normal development of stamens
Maturation of floral organs
Anthesis
Warm temperatures inhibit normal development of ovaries

plants began to deteriorate in growth and flower production due to inadequate size of the containers. All plants began to exhibit female sterility and later failed to produce any flowers. Under high temperatures, the plants degenerated even sooner than 30 weeks. The results of this study are presented graphically (Plate XII).

The plants of group one grown out-doors under natural conditions were extremely stable in sexual expression (Plate XII, Figs. 52, 53, and 54). Line 17CF6 (Fig. 52) showed a continuous production of Magnitude 2 (highly carpellodic) flowers, Line 17AF6 (Fig. 53) showed a continuous production of Magnitude 5 flowers (normal elongata type flowers), and Line 13BF6 (Fig. 54) showed a continuous production of Magnitude 7 (highly sterile) flowers.

Under the cool night temperatures of a reefer box all the plants of group two shifted toward carpellody (Plate XII, Figs. 55, 56, and 57). Line 17CF6 (highly carpellodic) shifted from Magnitude 2 to Magnitude 1 after 8 weeks, Line 17AF6 (low carpellody/low sterile) shifted from Magnitude 5 to Magnitude 3 after 8 weeks, and Line 13BF6 (highly sterile) shifted from Magnitude 7 to Magnitude 5 after 6 weeks. The 8 week period required for the sex change to appear in Lines 17CF6 and 17AF6 corresponds with the estimated period of 7 to 10 weeks from initiation of stamens to anthesis. Likewise, the 6 week period before a change occurred in Line 13BF6 (when only the ovary changed) confirms the estimated 2 week difference in time of initiation of stamens and ovary.

Lines 17CF6 and 17AF6 of the initial group three plants subjected to high day temperatures of approximately 105°F. stopped producing ovaries in about 4 weeks; stopped producing stamens after 2 more weeks, and died

PLATE XII

Figure 52. Response of high carpellodic line grown under normal outdoor conditions.

Figure 53. Response of low carpellodic/low sterile (normal) line grown under normal outdoor conditions.

Figure 54. Response of high sterile line grown under normal outdoor conditions.

Figure 55. Response of high carpellodic line subjected to cold night temperatures.

Figure 56. Response of low carpellodic/low sterile (normal) line subjected to cold night temperatures.

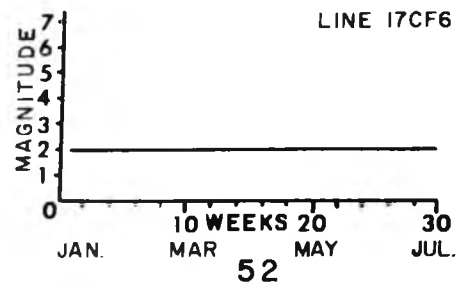
Figure 57. Response of high sterile line subjected to cold night temperatures.

Figure 58. Response of two groups of high carpellodic plants grown under high day temperatures.

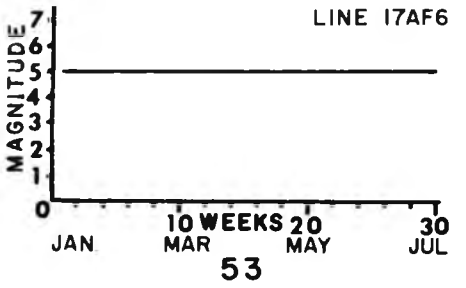
Figure 59. Response of two groups of low carpellodic/low sterile (normal) plants grown under high day temperatures.

Figure 60. Response of two groups of high sterile plants grown under high day temperatures.

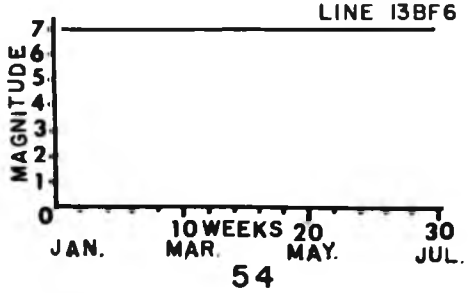
NORMAL OUTDOOR TEMPS.



52

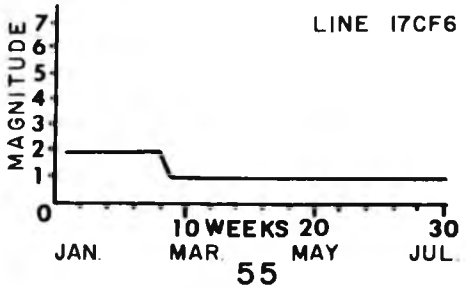


53



54

COLD NIGHT TEMPS.



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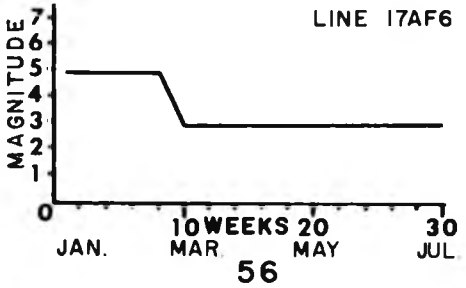

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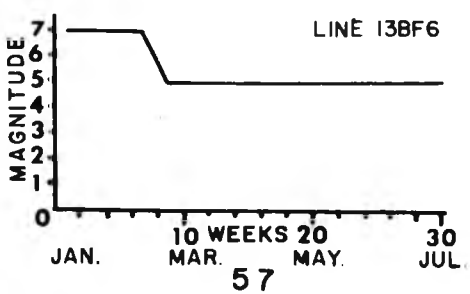
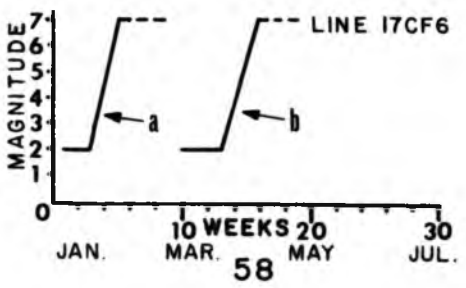


57

HIGH DAY TEMPS.



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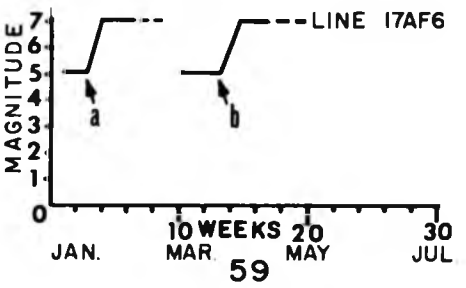

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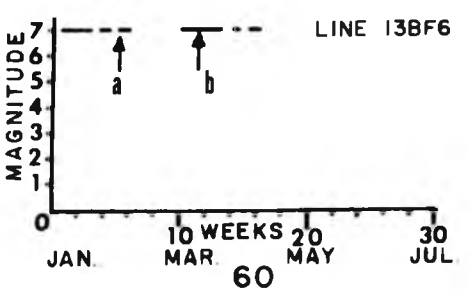


60

in 2 more weeks (Plate XII, Figs. 58 and 59). Line 13BF6 ceased flowering about 2 weeks after being placed in the glasshouse and died shortly afterward (Plate XII, Fig. 60).

A replacement group three plants was placed in the house at about 100°F. Once again, the plants soon exhibited female sterility and ceased flowering in 2 to 6 weeks, and died shortly afterward (Plate XII, Figs. 58, 59, and 60).

These inbred lines had previously been observed to be very stable in sexual expression under field conditions. These inbred lines placed in the different temperature regimes all showed a shift toward carpellody when exposed to a lower temperature and a rapid shift to female sterility when exposed to high temperatures. Furthermore, the time interval between the initial temperature exposure and the appearance of flowers showing a sex change was the same as the interval from initiation to anthesis determined from the morphological studies, and the 2 week difference between the initiation of stamens and ovaries was also observed in both studies. Thus it seems likely that stamens may be affected by changes in temperature occurring about 8 weeks prior to anthesis and ovaries may be affected by changes in temperature occurring about 2 weeks later or 6 weeks prior to anthesis.

C. CORRELATION BETWEEN 2 BASIC CLIMATIC FACTORS (TEMPERATURE AND RAINFALL) AND PROGENIES OF 3 INBRED LINES

In making the correlations of the class graphs (Plate I) with the temperature and rainfall graphs (Plate III), it was noticed that plants appeared to respond to changes in climate slightly differently

according to whether it was winter or summer and whether it was stamens or ovaries which were affected. For this reason, the correlations were carried out in a series of steps and the results charted (Table VII).

Since ovaries of carpellodic plants (Classes 1 through 5) do not change, and stamens of female sterile plants (Classes 7 through 11) do not change, no correlations were made for these organs in these plants.

Two general patterns emerge from the correlations; the developmental time required from initiation to anthesis appears to be about 1 to 2 weeks longer for all plants during the winter months as compared to the summer months, and, ovaries appear to exhibit sex shifts about 1 to 2 weeks sooner after temperature and rainfall changes occur than do the stamens. This pattern is present in all observed classes even though variations in length of time required to exhibit sex shifts ranged from a high of 10 weeks to a low of 4 weeks (Table VII). It appears that the time of highest susceptibility to climatic variations for the average plant would be 6 to 8 weeks prior to anthesis. Sexual changes were exhibited an average of 7 weeks prior to anthesis in the combined class made up of an average of all plants in the orchard.

The data presented thus far have established that temperatures which affect sex changes in papaya occur most often from 6 to 8 weeks prior to anthesis. Using this as a basis, the shifts in sex expression of each class in Figures 1 to 11 were compared to mean temperature changes occurring 7 weeks prior to anthesis (Plate XIII). Each class showed a different level of magnitudes and the shifts between magnitudes varied somewhat from class to class but the general pattern of changes in magnitude was consistent. That is, increases in temperature

TABLE VII. WEEK OF HIGHEST CORRELATION PRIOR TO ANTHESIS BETWEEN STAMENS AND OVARIES DURING SUMMER AND WINTER CONDITIONS AND DIFFERENCE BETWEEN TEMPERATURES (A), MAXIMUM TEMPERATURE (B), MINIMUM TEMPERATURE (C), AND RAINFALL (D)

CLASS	WINTER								SUMMER							
	STAMENS				OVARIES				STAMENS				OVARIES			
	A	B	C	D	A	B	C	D	A	B	C	D	A	B	C	D
1	7	10	10	10								8				
2	9	8	8	8					9	7	8	8				
3	8	9	8	8					8	8	7	7				
4	10	8	9	8					8	8	7	8				
5	10	8	8	10					8	6	7	7				
6	9	9	8	8	8	8	7	8	8			8	8	6	6	7
7					8	8	7	8					7	6	6	8
8					8	8	7	8					7	7	6	8
9					8	8	6	8					6	6	5	7
10					7	8	5	8					6	6	4	6
11					7	7	7	8								
all	9	8	8	10	7	7	7	7	9	7	7	7	7	6	6	7

PLATE XIII

Shift in magnitudes of Classes 1 through 11 in response to temperature changes 7 weeks prior to anthesis.

Figure 61. Class 1

Figure 62. Class 2

Figure 63. Class 3

Figure 64. Class 4

Figure 65. Class 5

Figure 66. Class 6

Figure 67. Class 7

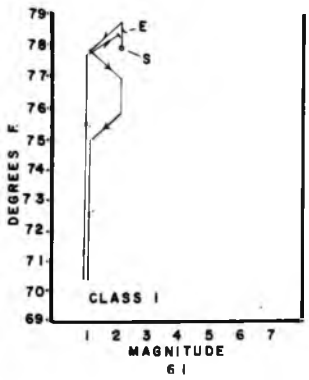
Figure 68. Class 8

Figure 69. Class 9

Figure 70. Class 10

Figure 71. Class 11

"S" - starting point of each chart
"E" - ending point of each chart
"A" - represents a point in March when average temperatures rose about 1°F. above the winter mean



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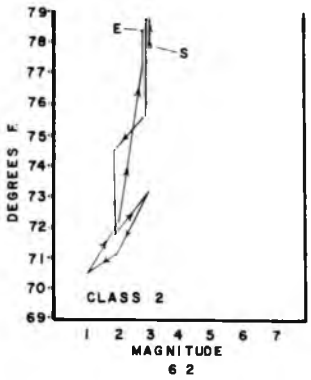

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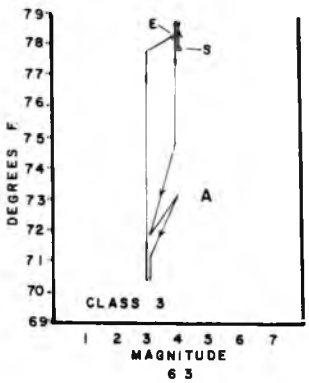

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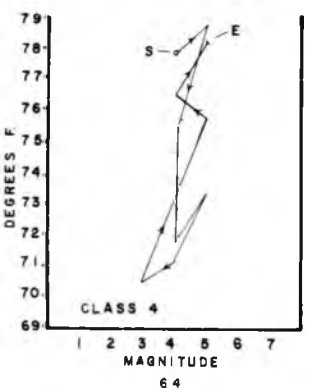

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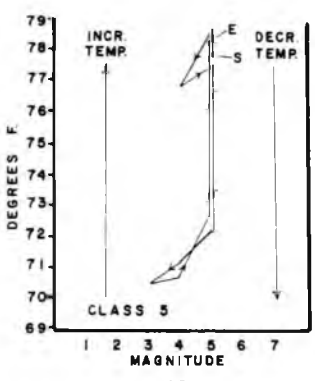

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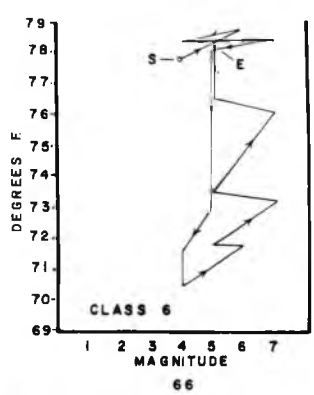

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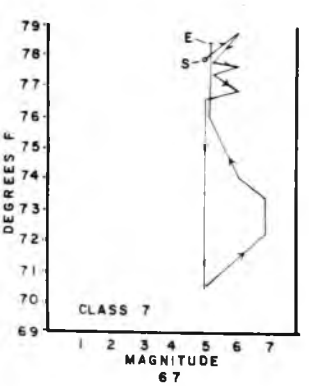

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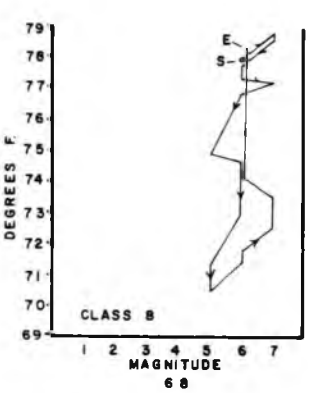

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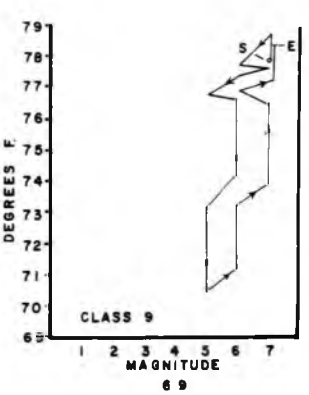

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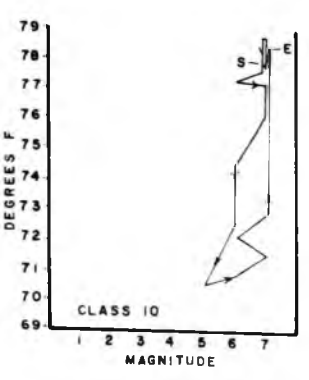

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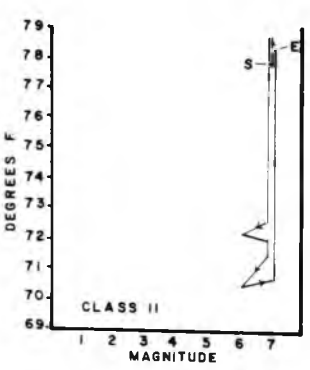

71

generally shifted magnitude (sex) toward female sterility and decreases in temperature generally shifted magnitude toward carpellody.

Generally, the minimum and maximum temperatures paralleled each other closely; that is, when one temperature was high, the other tended to be high also and when one was low, the other was also low. With the onset of the winter cycle in November, average weekly temperatures began to decline at an average rate of about 0.7°F. per week over a 10 week period. Temperatures remained at the winter low (mean 70.5°F.) for about 6 weeks. At that point, in March, a general increase in average temperature (about 1°F.) occurred which lasted for about 3 weeks. Temperatures then returned to their previous winter low where they remained for 3 more weeks. Around mid-April, temperatures began increasing at the rate of approximately 1°F. per week over a 7 week period. By June, temperatures reached their maximum for the summer period (mean 77.5°F.).

In Plate XIII, a horizontal line means there was no temperature change but a shift in magnitude of sex expression occurred. Conversely, if there was a temperature change but no change in the sexual expression, there is a vertical line on the graph. If there was a temperature change and a shift in magnitude, the line would slant to the right or left depending on the amount of change and the direction of change. There is no time sequence present in the charts (Plate XIII) but simply a sequence of how much the temperature varied up or down and the response of sex expression to those changes.

The charts (Plate XIII) represent the response of plants within each class to the temperature changes and thus, the apparent ease or difficulty

of causing shifts in magnitude. This relationship between magnitude of sex shift and temperature change can be more easily seen by discussing each class individually.

Class 1 (Fig. 61) is made up of highly stable carpellodic plants, mostly from parental line 17CF6, but also including any other plants that showed sex expression of Class 1 type. The initial rise of 0.7°F. in temperature caused no shift in magnitude in Class 1 as indicated by the vertical line proceeding up from the starting point ("S") on the chart. After this, the average temperature began to decrease at the rate of 0.7°F. per week and an almost immediate change occurred shifting the plants to a higher degree of carpellody (Magnitude 2 to Magnitude 1). However, even though temperatures were still decreasing at the same rate, sex expression shifted back to the less carpellodic state (Magnitude 2) within a short time. Continued temperature decreases resulted in a shift in sex back to Magnitude 1. The plants remained in Magnitude 1 where no further shift can occur for approximately 5 weeks although there was a 4°F. decrease during that period. With the onset of summer it took a temperature rise of nearly 7°F. before the plants became less carpellodic (Magnitude 2). The chart for this class shows the relative stability of these plants in their sexual expression.

Class 2 (high carpellody - Fig. 62) began in Magnitude 3 and the initial 0.7°F. rise in temperature had no influence on sexual expression. However, as soon as temperatures began to decline with the onset of winter at a rate of about 0.7°F. per week, magnitude of sex expression shifted towards more intense carpellody. This shift was rather gradual over a span of magnitude (3 down to 2) over a temperature

drop of about 4°F. At this rate, the plants of Class 2 required about 4°F. of temperature drop to shift one magnitude towards carpellody. A further temperature drop of 2.5°F., however, resulted in no further shift towards carpellody. The sudden increase in temperature by 1°F. in March over a period of 3 weeks effected a shift in sex expression from Magnitude 2 to 3. When temperatures decreased to their previous level, sex expression returned to Magnitude 2. Further decreases in temperature (0.8°F. more) to the lowest level of the year (70.5°F.) effected additional change in sex expression to Magnitude 1, the highest level of carpellody. The general and steady increase in temperature as summer approached (1°F. per week for about 8 weeks) resulted in a gradual steady shift in sex expression back to Magnitude 3. This indicated that a temperature increase of somewhat more than 3°F. was necessary to cause a shift in sex expression of one magnitude and for each 3°F. rise in temperature, the plants' sex shifted to the next higher magnitude. The range of magnitudes exhibited in Class 2 was relatively narrow, but compared to Class 1, the plants tended to show a more direct response to the variations in temperatures encountered.

Class 3 (moderate carpellody - Fig. 63) showed a pattern similar to that of Class 2 even in March when a short 1°F. rise in average temperature induced a shift from Magnitude 3 to 4 indicated by the spur-like protrusion at point "A" on the Class 3 chart (Plate XIII, Fig. 63). This class seemed to resist shifts away from Magnitude 3. The flowers of Magnitude 3 are commonly called "pentandria" and are referred to by some workers as a "normal" type flower (34, 97). The resistance to shift from this level appears to support the theory that the pentandria flower represents a genetically stable type. This resistance

to change was illustrated by the relatively large fluctuations in temperatures required to effect a shift from one magnitude to the next.

Class 4 (slight carpellody - Fig. 64) is representative of a class which is highly responsive to temperature changes. As shown in the chart (Plate XIII, Fig. 64), there were only a few times when a temperature change did not result in a shift in magnitude of sex expression. None of these shifts were sudden or of a large degree. Each showed a steady response at the rate of about one magnitude of shift for every 1° to 3° F. change in temperature. This is indicative of an additive effect of temperature on the expression of genes which affect sex changes.

Class 5 (very slight carpellody - Fig. 65) is very close to the highly stable normal type (elongata flowers), but slight amounts of carpellody may occur (Table III). This stability of Magnitude 5 flowers was indicated by the large decreases or increases in temperature necessary to force a shift in sex. This class of plants required about a 5°F. drop in temperature before a change to a lower magnitude occurred. After this point was reached, additional decreases in temperature (about 2°F. more) effected a steady shift toward increasing carpellody (Magnitude 5 to 3). This shift was also reversible in the same way. As soon as temperatures began to rise, sex expression shifted back toward Magnitude 5. Small 1° or 2°F. temperature changes caused little or no effect on this class. Class 5 tended to give the impression that a threshold value of genetic resistance to shift in sex may be present in the plants. In classes such as 2, 4, 7, 8, and 9, this threshold value may be low and shifts in sex occur easily in response to slight variations in temperature or other external factors. In other classes, such as 1, 3,

5, 6, 10, and 11, the threshold value appears to be higher and greater external variation must occur before plants respond.

Class 6 (Fig. 66), an essentially normal class exhibiting shifts to sterility or carpellody of short duration, showed a high degree of sensitivity to changes towards sterility. Changes in sex expression in this class were always of very short duration, never lasting more than 3 weeks at a time and for no more than 12 weeks during the year. The initial 0.7°F. temperature increase resulted in a sex shift from Magnitude 5 to 7 (increasing sterility). As the temperature began decreasing with the onset of winter, the sex almost immediately shifted to a normal sex type (Magnitude 5) where it remained throughout a 6°F. drop in temperature. The slight 1°F. increase in temperature for 3 weeks in March resulted in a sex shift to Magnitude 7. A reduction of this temperature shifted the sex back to Magnitude 5. A further decrease of 1°F. to the lowest temperature of the year (70.5°F.), resulted in a shift to Magnitude 4. As temperatures began increasing with the onset of summer, female sterility rose to Magnitude 7. Sex quickly shifted back to Magnitude 5 but appeared to fluctuate back and forth between Magnitudes 5 and 7 throughout the rise in temperatures to the high summer level. In this case, sexual shifts followed closely the pattern of temperature changes as long as temperatures were decreasing. Plants seemed to respond quickly to rising temperatures, however. The plants were placed in Class 6 because they exhibited normal flowers more than 90 per cent of the time and showed sterility or carpellody for only a limited time. At this point, no explanation of the strongly responsive actions can be made except to note that the threshold value for resistance to shift

toward female sterility must be very low.

Class 7 (very slightly sterile - Fig. 67) showed definite response to temperature changes. The initial temperature rise of 0.7°F. shifted sex expression from Magnitude 5 to 6. When the temperature decreased, the plants returned to Magnitude 5 and remained there in spite of a drop of nearly 6°F. Plants that exhibited sterility in Classes 7 to 11 showed strong resistance to shift towards carpellody under conditions encountered at Malama-Ki. An increase in temperature of about 1°F. during March caused Class 7 plants to shift in sex from Magnitude 5 to Magnitude 6 but quickly returned to Magnitude 5 as the temperature decreased again. When the temperature began rising as summer approached, the sex expression of the plants in this class shifted to Magnitude 6. The plants remained in Magnitude 6 in spite of a rise of more than 3.5°F., a level nearly approaching the maximum temperature encountered near the end of the year cycle. At this point, with temperatures stable at their highest point, sex expression shifted from Magnitude 6 to 5. The sudden reversion from a nearly completely sterile state to fertility without any corresponding change in temperature may be due to other factors not measured in these charts.

Performance of the plants in Class 8 (slightly sterile, Fig. 68) was similar to those of Class 7. The plants of Class 8 initially exhibited sex expression of Magnitude 6 and remained there with a 0.7°F. rise in temperature. They gradually shifted to Magnitude 5 as temperatures dropped about 3°F. The shift to Magnitude 7 in response to rising temperatures after the winter season was slightly faster and lasted longer in Class 7. These plants also returned from Magnitude 7 to 6 near the

end of the cycle with no apparent change in temperature.

The plants of Class 9 (moderately sterile, Fig. 69) began with a sex expression of Magnitude 6 and shifted to Magnitude 7 as the temperature increased by 0.7°F. As the temperature dropped with the onset of winter, the sex shifted gradually to Magnitude 5. The degree of shift in sex was about one magnitude toward decreasing sterility for each 3°F. drop in temperature and was constant over the entire range encountered. As temperatures began to rise after the winter period, sex expression showed increasing sterility, shifting from Magnitude 5 to 7 at about the same rate as it fell earlier in the year in response to decreasing temperatures. The plants became less sterile by one magnitude near the end of the cycle as the temperature neared its highest point, but they returned to Magnitude 7 at the end of the cycle. The type of response exhibited by Class 9 seems to indicate strongly a direct relationship between sex and temperature. That is, the greater the mean temperature increase or decrease, the greater the proportionate shift in magnitudes. The threshold of sensitivity to temperature appears to be very low as sex changes occurred freely with small or steady changes in temperature.

Class 10 plants (highly sterile, Fig. 70) started their cycle in Magnitude 7. The first 0.7°F. increase in temperature had no effect on sex change since the plants were already at the maximum level of sterility. A decrease in temperature of nearly 3°F. at the onset of winter resulted in no sex shift. However, further steady decreases in temperature of about 4°F. resulted in a steady shift in sex expression to Magnitude 5. This point was not reached until the lowest temperature

was encountered. As soon as the temperature began to increase with the onset of summer, sex expression shifted back to Magnitude 7. The plants reached Magnitude 7 rapidly at the rate of about one magnitude of shift for each degree rise in mean temperature and remained there for the rest of the cycle. These plants apparently exhibited a moderately high threshold level of sensitivity requiring a minimum of at least a 3°F. drop in temperature before shifting to lesser degrees of sterility. Temperature drops greater than 3°F. caused sex shifts toward lesser sterility which appeared to be additive. The shift in magnitudes (up or down) appeared to be directly proportional to changes in degrees of temperature after the threshold of sensitivity was overcome.

Class 11 (Fig. 71) represents the strongest possible sterility and contains plants of the parental line (13BF6) as well as a few other trees exhibiting similar sex type. The reaction of these plants to mean temperature variations was virtually identical to that of Class 10 except that here the plant's threshold value was higher, requiring a full 6°F drop in temperature before a shift to lesser sterility was observed. After this point was reached, additional decreases in mean temperature caused sex expression to shift toward increasing fertility (Magnitude 7 to 6). As mean temperatures rose again with the onset of summer, sterility increased to that represented by Magnitude 7. Again, changes in sex expression after the threshold value was overcome appeared to be additive.

A general pattern of sex shifts in relation to mean temperature changes was clearly manifested by the plants in the 11 classes. Plants exhibited threshold values for sensitivity to temperatures, either low and easily susceptible to changes in temperature or high and resistant to change. The level of the threshold value was apparently dependent upon

the genetic characteristics of the plant involved. Parental lines (17AF6, 17CF6, and 13BF6) represented mostly in Classes 1, 6, and 11, respectively, were bred and selected for stability of sex characteristics under the environmental conditions encountered. Therefore, a high threshold value was developed. However, the threshold value appeared to change as outcrossing occurred. Generally, when the temperature representing the threshold value was exceeded, an apparent direct relationship between temperature and sex was encountered in nearly all cases.

Two other occurrences were also noted. First, the plants in classes showing varying degrees of sterility (7 to 11) showed relatively greater ease in shifting sex expression than did the more carpellodic classes (1 to 5). That is, a given mean temperature variation caused a stronger response of the plants in the sterile magnitudes than in the carpellodic ones. The possibility that greater morphological changes in plant organs are necessary to shift from one magnitude to the next in the carpellodic magnitudes than in the sterile magnitudes cannot be ignored. This seems reasonable inasmuch as it appears to involve less of a morphological change to reduce or increase the number of carpels in a normally 5 carpellate, single celled ovary than it is to transform as many as 10 stamens into ovary-like structures. Secondly, it appeared that with increasing threshold levels of sensitivity, a greater intensity of environmental change became necessary to effect shifts in sex expression. Short sudden changes in climate no longer effected sexual shifts in plants with high threshold values as they apparently did in plants with low threshold values. The possibility exists that plants

in the carpellodic classes have higher inherent threshold values than those in sterile classes.

C. GENETICS AND INHERITANCE OF THE FACTORS FOR CARPELLODY AND STERILITY

Crosses were made in all combinations between the highly carpellodic inbred line (17CF6), the female sterile inbred line (13BF6), and the inbred normal line (17AF6) in order to study the mode of inheritance of carpellody and sterility. All plants for these genetic studies were grown and evaluated at the same time in the field at Malama-Ki.

Cross between the high carpellody line (17CF6) and the normal line (17AF6)

The frequencies of individuals in the different classes for the parents, F_1, F_2, and BCP_2 are presented in Table VIII and Figure 72. Although the 2 parents had been inbred for 6 generations, there were still some variability present. Variability in these inbred lines indicate the presence of some residual heterozygosity and the highly sensitive nature of this character to environmental changes. Although all plants were grown at the same time in the same field, there may well have been microclimatic differences within the field.

An F_1 of 2 homozygous parents is expected to be genetically uniform and exhibit low variability, about equal to that of the parents. The F_1 of this cross (Fig. 72) showed variability slightly higher than either of the parents, indicating some degree of segregation of characters and that 6 generations of inbreeding may not be enough to bring all quantitative characters within the parents to homozygosity. The

PLATE XIV

Figure 72. Segregation pattern of the parents and progenies of the cross High Carpellody X Normal.

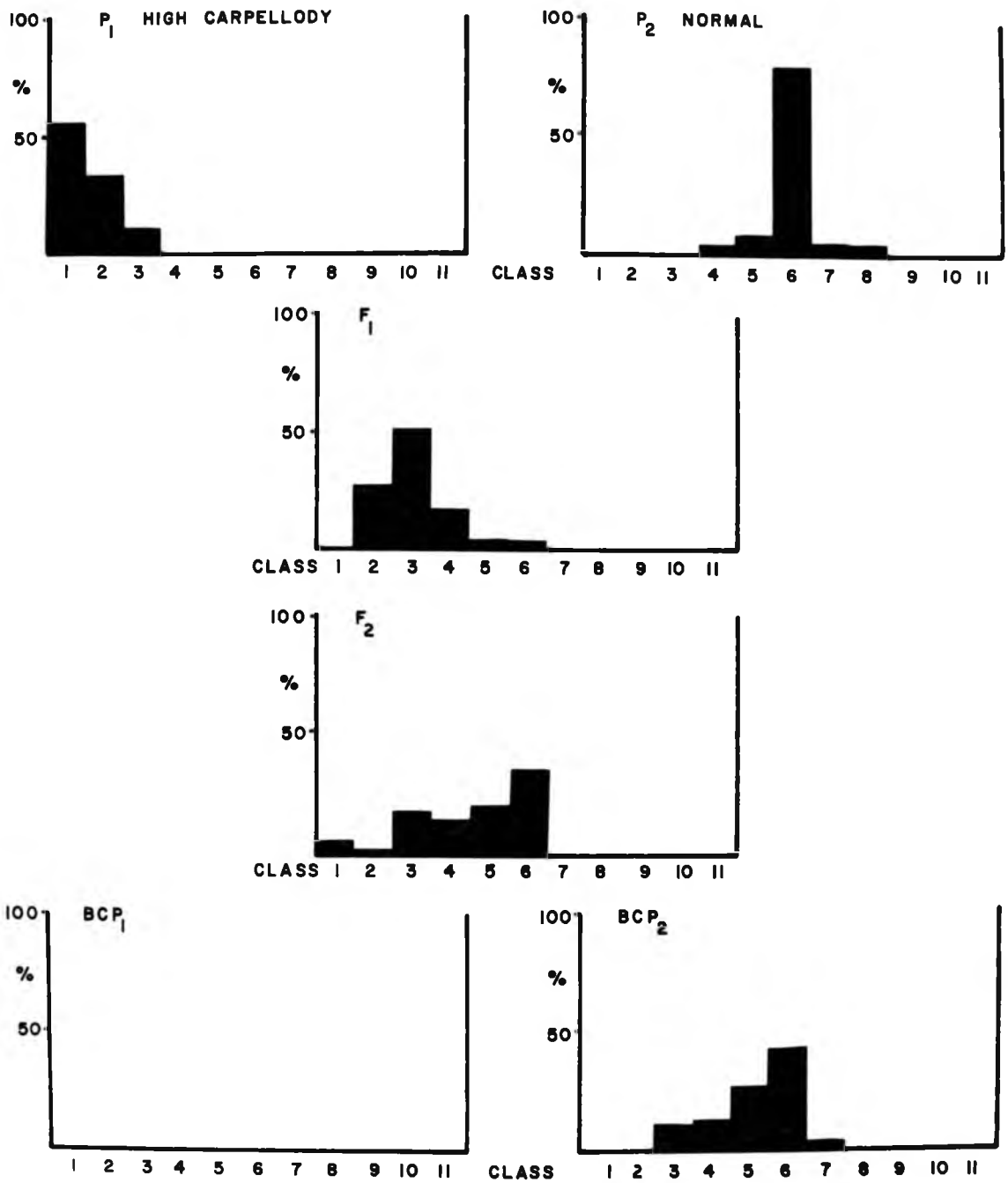
100
%
50
P1 HIGH CARPELLODY
1 2 3 4 5 6 7 8 9 10 11
100
%
50
P2 NORMAL
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
F1
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
F2
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
BCP1
1 2 3 4 5 6 7 8 9 10 11
100
%
50
BCP2
CLASS 1 2 3 4 5 6 7 8 9 10 11

TABLE VIII. FREQUENCY DISTRIBUTIONS FOR SEX CLASS IN THE PARENTS, F_1, F_2, AND BCP_2 GENERATIONS IN THE CROSS HIGH CARPELLODY X NORMAL

FAMILY	CLASS											N	$\overline{X}$ ± S.E.	Theoretical means		TOTAL VARIANCE
	1	2	3	4	5	6	7	8	9	10	11			ARITH.	GEOM.	
P_1	5	3	1									9	1.56 ± .323	3.79	3.06	5.56
P_2				1	6	54	3	3				67	6.01 ± .152			17.55
F_1		8	15	5	1	1						30	3.07 ± .494			19.12
F_2	3	1	7	6	8	14						39	4.46 ± .606			51.70
BCP_1												0		2.68	2.19	
BCP_2			4	5	11	17	2					39	5.20 ± .658	4.90	4.29	34.20

variability of the F_2 is expected to be wide, since genetic variability as well as environmental variability is expected to be present. This F_2 population included all the classes of P_1 and all but the 2 uppermost classes of P_2 (Classes 7 and 8). The failure to recover the 2 P_2 classes and the highly uniform frequencies in each of the classes indicate that this F_2 population was not large enough to show the full range of possible classes. The variability of a backcross is expected to be intermediate between the F_2 and the parents and F_1. However, this backcross showed little loss of variability below that of the F_2, indicating that quite a large number of genes were probably involved here.

The arithmetic midpoint of the 2 parents was 3.79 and the geometric mean was 3.06. The observed F_1 mean (3.07) closely approximated the geometric mean. The F_2 mean (4.46), however, was closer to the arithmetic mean. This could indicate some degree of dominance, but is more likely due to an insufficient number of individuals in the segregating F_2 population. The backcross mean (5.20) was closer to the arithmetic mean (4.90) than to the geometric mean (4.29) for that cross. This again may indicate a lack of sufficient number in this progeny.

The variances of the parents, F_1, F_2, and BCP_2 are presented in Table VIII. The variance of P_2 is higher than that of P_1 and may be explained by the fact that the P_2 population was much larger than the P_1 population and covered a wider range. The variance of the backcross was lower than that of the F_2 as expected and intermediate between the F_2 and F_1 and parents. Variances of the F_2 and backcross were higher than those of the F_1 or the parents as expected in segregating

populations indicating the probable involvement of several genes and a lack of complementary genetic action.

Cross between the normal line (17AF6) and the high female sterile line (13BF6)

The results of this cross are given in Table IX and Figure 73. The high sterile line (13BF6) had been inbred for 6 generations like the other parental lines but still showed a small amount of variability. The variability would be due to environmental effects and the necessity to select a plant that is capable of producing a few normal flowers to perpetuate the line. Plants which remained constantly in Magnitude 7 never produced any seed. For this reason and also because reciprocal crosses showed no differences, the high sterile line was used as the male parent.

The variability of the F_1 was low, as was expected, but the mean (6.16) was shifted significantly towards the normal parent. This shift of the F_1 mean could indicate a high degree of dominance for the genes for normal, non-sterile flowers.

The F_2 population was highly variable and had a wide distribution range covering nearly all of the classes present in both parental lines. The lowest class of P_1 (Class 4) was not recovered. The F_2 mean approximated the geometric midpoint value of the parents rather than shifting towards the low sterile parent. This shift would have been expected if P_1 (normal) possessed dominant genes for normal sex expression.

The BCP_2 (high sterile) progeny recovered all of the classes present in the F_1 and the P_2. Its variability was slightly smaller than

PLATE XV

Figure 73. Segregation pattern of the parents and progenies of the cross Normal X High Sterile.

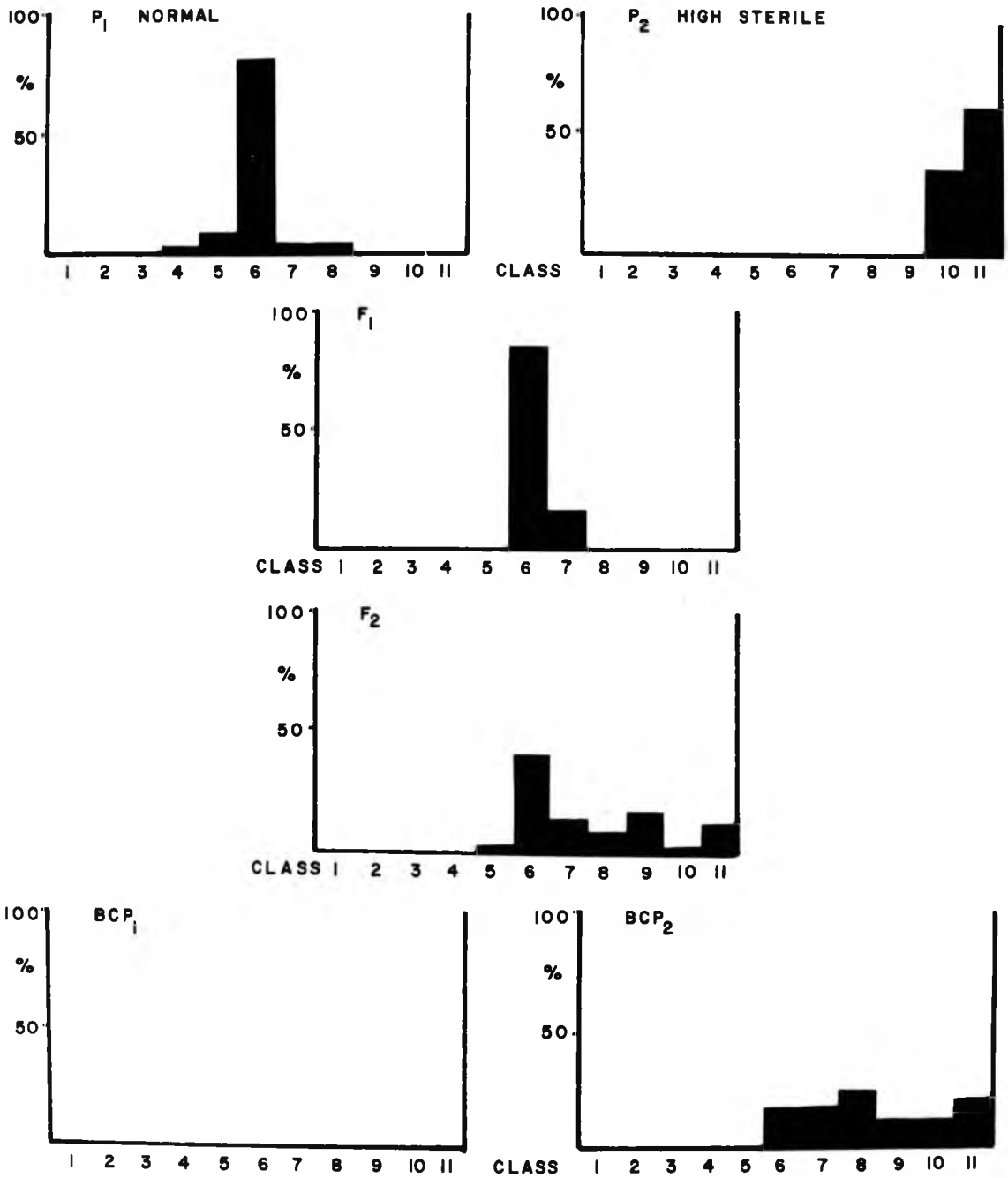
100
%
50
P1 NORMAL
1 2 3 4 5 6 7 8 9 10 11
100
%
50
P2 HIGH STERILE
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
F1
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
F2
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
BCP1
1 2 3 4 5 6 7 8 9 10 11
100
%
50
BCP2
CLASS 1 2 3 4 5 6 7 8 9 10 11

TABLE IX. FREQUENCY DISTRIBUTIONS FOR SEX CLASS IN THE PARENTS, F_1, F_2, AND BCP_2 GENERATIONS IN THE CROSS NORMAL X HIGH STERILE

FAMILY	CLASS											N	$\bar{X}$ ± S.E.	Theoretical means		TOTAL VARIANCE
	1	2	3	4	5	6	7	8	9	10	11			ARITH.	GEOM.	
P_1				1	6	54	3	3				67	6.01 ± .410	8.32	7.99	17.55
P_2										4	7	11	10.63 ± .281			5.11
F_1						26	5						6.16 ± .159			8.36
F_2					1	17	6	4	7	1	5	41	7.53 ± .859			63.71
BCP_1												0		7.17	6.93	
BCP_2						6	6	8	4	4	7	35	8.42 ± .718	9.48	9.21	53.10

the F_2 as expected and its mean (8.42) was shifted towards low sterility. Seeds for the backcross to the normal parent (BCP_1) did not germinate.

In the absence of dominance, the F_2 mean is expected to approximate that of the F_1. However, in this case, the F_2 mean (7.53) was significantly shifted toward the high sterile parent away from the F_1 mean (6.16). If this were a true representation of the inheritance pattern of this cross, then dominance for factors causing low sterility (more femaleness or a normal type) was exhibited. The same pattern of dominance of more femaleness was present in the high carpellody X normal cross but to a lesser extent (Plate XIV, Table VIII).

The variance of P_1 was found to be higher than that of P_2 as expected in view of the wider distribution range of P_1 as compared to the relatively narrow range of P_2. The variance of the F_1 was relatively low as expected in a non-segregating progeny. Also the variances of both the F_2 and the backcross were higher relative to the P_1, P_2, and F_1 as expected in segregating populations.

Cross between the high carpellody line (17CF6) and the high sterile line (13BF6)

The results of this cross are presented in Table X and Plate XVI. The F_1 showed a much wider distribution than either of the parents resembling that of the inbred normal line (17AF6). The mean (6.40) was very near the midpoint of the 2 parental lines (6.10) and the mean of the normal inbred line (6.01). It seems reasonable to conclude that this is the distribution range that is obtained from a non-segregating population with a mean of about 6.

PLATE XVI

Figure 74. Segregation pattern of the parents and progenies of the cross High Carpellody X High Sterile.

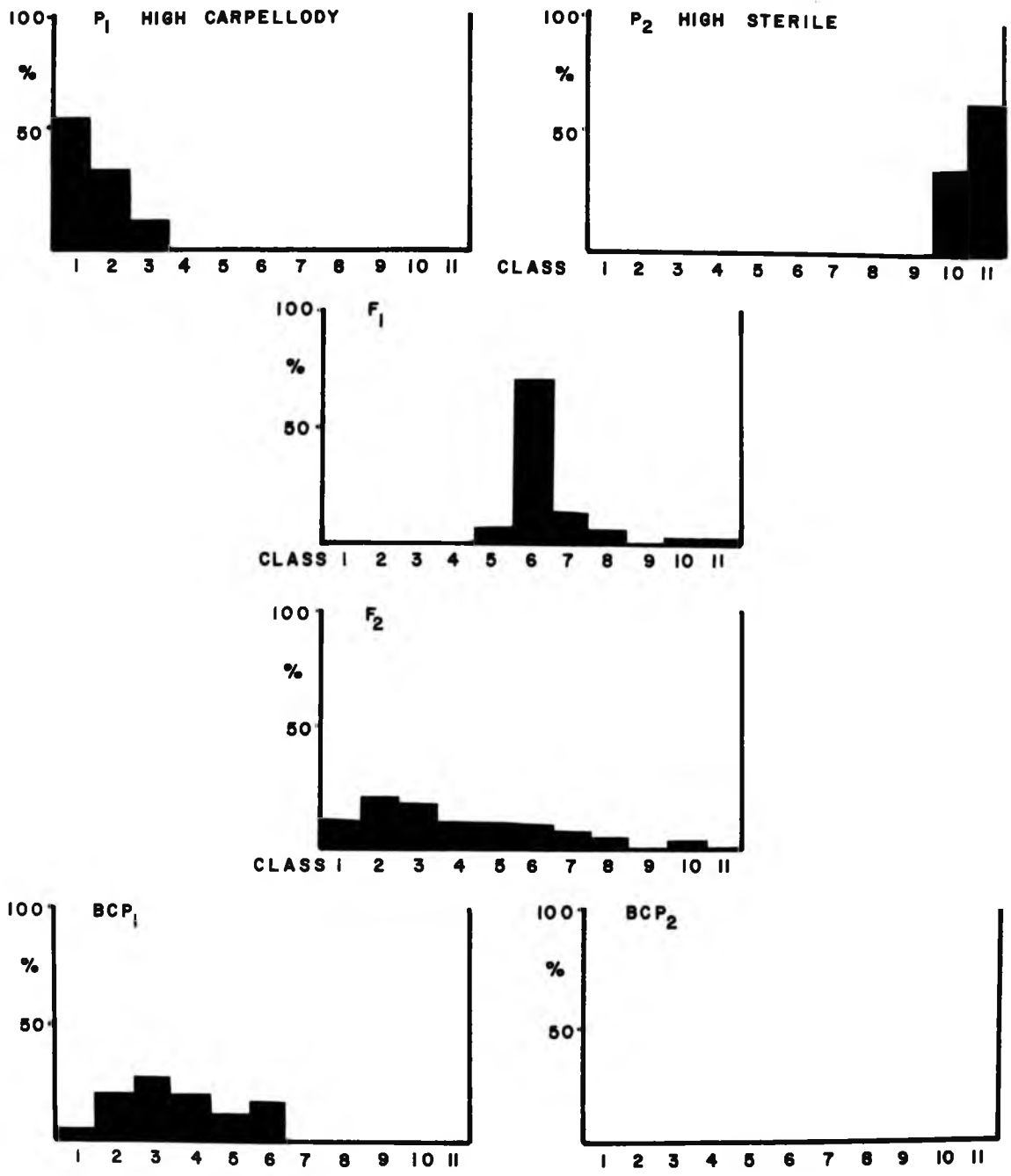
100
%
50
P1 HIGH CARPELLODY
1 2 3 4 5 6 7 8 9 10 11
100
%
50
P2 HIGH STERILE
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
F1
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
F2
CLASS 1 2 3 4 5 6 7 8 9 10 11
100
%
50
BCP1
1 2 3 4 5 6 7 8 9 10 11
100
%
50
BCP2
1 2 3 4 5 6 7 8 9 10 11

TABLE X. FREQUENCY DISTRIBUTIONS FOR SEX CLASS IN THE PARENTS F_1, F_2, AND BCP_1 GENERATIONS IN THE CROSS HIGH CARPELLODY X HIGH STERILE

FAMILY	CLASS											N	$\bar{X}$ ± S.E.	Theoretical means		TOTAL VARIANCE
	1	2	3	4	5	6	7	8	9	10	11			ARITH.	GEOM.	
P_1	5	3	1									9	1.56 ± .323	6.10	4.07	5.56
P_2										4	7	11	10.63 ± .231			5.11
F_1					3	30	6	3		1	1	44	6.40 ± .954			32.80
F_2	4	8	7	4	4	4	3	2		1		37	4.00 ± 2.22			70.00
BCP_1	1	5	7	5	3	4						25	3.64 ± .734	3.83	2.52	30.64
BCP_2												0		8.37	6.57	

The F_2 population of this cross showed a wide distribution and included all but 2 sterile classes (9 and 11). Considerable skewness towards carpellody was apparent both in the distribution and in the mean (4.00). This shift of the F_2 cannot be interpreted to indicate dominance, since any dominance would have been indicated by a shift in the F_1 mean toward the parental line with the dominant genes, and a shift in the F_2 away from the F_1 toward the recessive. The most plausible explanation for the skewness observed here is that insufficient numbers of F_2 plants were grown to accurately represent the F_2 distribution.

The backcross (BCP_1) to the high carpellody parent showed a relatively large variability, but less than that of the F_2. The mean (3.64) was shifted slightly, but not significantly toward carpellody.

The variances for P_1 and P_2 were quite low as expected of inbred lines. The variance for the F_1 was higher than the parental variances indicating some degree of segregation, probably from impure parents. The variance for the F_2 was high as expected of a segregating population. Variance for the backcross was considerably lower than that for the F_2 indicating a form of gene action not observed in the crosses low carpellody X high carpellody and low sterile X high sterile, perhaps epistatic.

The data for the three crosses indicates partial dominance for the factors for carpellody. It is also strongly indicative that many sets of genes are involved in sexual expression. It appears, on the basis of the histograms and means and variances in the various tables, that inheritance of the factors for carpellody and sterility is quantitative with multiple factors involved. No pattern of simple mono-factorial

inheritance was observed. The mean of the F_1 was found to deviate significantly from mid-parental values in the crosses high carpellody X normal and normal X high sterile. The F_1 did not deviate significantly from the mid-parental value in the high carpellody X high sterile cross. There were evidence of complete dominance in the cross normal X high sterile and partial dominance toward carpellody was noted in other crosses. Significant deviation of the F_1 means from the arithmetical midpoint values of the parents was generally removed or reduced when compared to the geometric mean, indicating at least partial dominance of the factors for carpellody.

There appears to be little consistency in the deviations of the F_2 progenies from the calculated means of the parents and the F_1 means. The distribution range of the F_2's recovered a high portion of the parental range but did not recover all the classes. This may be attributed to insufficient sample size in the F_2 or to the presence of a relatively large number of pairs of factors affecting the characters carpellody and sterility.

Generally, the data presented seem to indicate the presence of many genetic factors controlling the sexual expression of the plant. Some of these factors have been referred to as "suppressors" of one sex or the other (45, 99, 100) and interest has been shown in "modifying factors" of sex expression (2, 21, 26, 33, 39, 40, 41, 44, 45, 47, 48, 68, 79, 94, 99, 100, 115, 116, 117).

Storey (99) listed hypothetical modifying sex genotypes for the principal sex types, male, hermaphrodite, and female. The only difference between the male and hermaphrodite sex types was hypothesized

by the presence of "mp", the male peduncle characteristic in the male genotype. He also postulated the presence of "sa", a suppressor of the androecium present in both the male and hermaphrodite, and "sg", a suppressor of the gynoecium present in both the male and hermaphrodite. In this case, the male and hermaphrodite are of the same genetic makeup except for the peduncle character. Hamilton and Izuno (31) also stated this. Fruitful and completely unfruitful male types have been reported. On the basis of this information, it could be assumed that differences in the phenotypic sexual expression of the papaya may be due solely to the action of "sa" and "sg" genes. These genetic factors may be homozygous as in the completely female-sterile male which never shows sex changes or heterozygous as in the fruiting male and the sex changing hermaphrodite. Completely fruitful hermaphrodites may be due to the lack of both of the alleles "sa" and "sg" (99). The exact nature of these two genetic characters is not known. It is entirely possible that they may be multiple factors. This is borne out by the quantitative nature of sexual expression exhibited by hermaphrodites with changes in temperature. The presence of threshold values in some cases may also indicate that a numerical value can be assigned to the quantity of factors "sa" and "sg". It may be proposed that these factors are normally in a heterozygous state which would allow for the apprent large degrees of sexual variability in outcrossed plants. With continuous inbreeding which leads to homozygosity of characters within the plant, there appears to be a buildup of threshold value which must be overcome before sexual variations can occur.

Storey's "sg" factor (99) when suppressed, would allow for normal development of an ovary, and when activated, would suppress the development of a normal ovary, either completely or partially. However, the "sa" factor may not operate in the same manner. If the "sa" factor suppressed the development of normal stamens, the stamens do not disappear or become vestigial as the ovary does, but rather takes on female characteristics. It appears that "sa" factors may add female characteristics in the development of male organs and "sg" factors simply subtract female characteristics from female organs.

Hofmeyr (44) postulated the existence of various modifying factors for his dioecious lines in South Africa. The data obtained in this study seem to support the adaptation of his proposed genotypes to the hermaphroditic lines used. Hofmeyr's genotype indicates the presence of two equal halves of one group of genes; one half containing genes effective in altering the phenotype of the major male genotype ("M_1m") and the other half effective in altering the phenotype of the major female genotype ("mm"). In a heterozygous state, the genes are ineffective in changing the sex, but when the proper set (first half or second half) becomes homozygous, changes in sex occur. It would not be unreasonable, on the basis of this study, to assume these same types of modifying factors to be present in the hermaphroditic lines used here except that the number of genes present would be many more than indicated by Hofmeyr to account for the almost continuous range of sex types (phenotypes) observed. Also, the genotype would no longer contain an equal proportion of genes for carpellody and sterility, but rather would show about two thirds for carpellody and one third for sterility.

Since all plants in this study were grown in one location at the same time, any variation in sex patterns from plant to plant could be considered due to genetic or genetic-environmental interactions. Environmental variance would be about the same for every plant. Large numbers of modifying factors inducing carpellody could make the "M_2m" genotype exhibit a phenotype like that of the high carpellody line (17CF6). A large number of modifying factors inducing sterility could make the "M_2m" genotype exhibit a phenotype like that of the high sterile line (13BF6).

Storey (99) postulated the existence of "sa" and "sg" genes and that they were two independent sets of genes. He also noted that some epistasis between the factors for carpellody and sterility may be present.

The genetic data gathered in this study can be used to expand the theories of Storey and Hofmeyr to give a hypothetical model of the principal modifying factors of sex expression. It is clear that a normal di-hybrid type model will not satisfy the ratios obtained in the progenies of all three crosses in this study.

A tri-hybrid model represents the least number of factors possible to fit the field data, and it is possible that many more modifying factors may be present which may further alter the ratios. The model consists of 3 loci, 2 for carpellody (c/c c/c) and 1 for sterility (s/s). These factors are not linked and act independently. Their normal alleles (+) are partially or completely dominant; i.e., in s/+, the + is completely dominant to the s, and in c/+, the + is partially dominant over c. Also, the s factor, whether homozygous or not (s/-) is epistatic over the c allele when the carpellody factors are heterozygous. If

either of the c factors is homozygous (c/c), the combined strength of the two alleles at one locus will overcome epistasis and carpellody will be exhibited. Table XI gives the genotypes and the corresponding sex types (phenotypes) which would be exhibited for each genotype.

When the genotypes of Table XI are applied to actual field data obtained from the crosses at Malama-Ki, the theoretical ratios fit very closely with those actually obtained (Table XII). Chi-square (X^2) in the F_2's and backcrosses in the crosses High Carpellody X Normal and Normal X High Sterile (Table XII) indicate a close fit between calculated and observed sex types. X^2 for the F_2 in the cross High Carpellody X High Sterile ($P = .14$) was acceptable ($P > .05$) and because of the small population in the F_2 involving segregation of many sex types, it is felt that the observed types are in line with the calculated ratio. X^2 for the backcross (BCP_1) in this cross yielded a p value of only .01 which indicates a poor fit. However, extremely small size of the BC population (25) may account for failure to recover many sex types, especially the High Carpellody types as many of these died during the experimental period and were not recorded.

It would appear, then, that Storey's hypothetical sex factors "sa" and "sg" can be represented by "c/c c/c" and "s/s", respectively, and further, that the epistasis he postulated is actually present under some conditions. Further crosses involving all stable sex types (High Carpellody, Pentandria, Normal, and High Sterile) should be made to support the model presented in Table XI.

TABLE XI. GENOTYPES OF MODIFYING FACTORS AFFECTING SEX EXPRESSION AND THE PHENOTYPES EXPECTED BASED ON THREE PAIRS OF GENES[a]

FREQUENCY	GENOTYPE	PHENOTYPES EXPECTED
1	c/c c/c s/s	High carpellody of Class 1 or 2
2	c/c c/c s/+	High carpellody of Class 1
1	c/c c/c +/+	High carpellody of Class 1, used as high carp parental type
4	c/c c/+ s/+	High carpellody of Class 2
2	c/c c/+ +/+	High carpellody of Class 1 or 2
4	c/+ c/c s/+	High carpellody of Class 2
2	c/+ c/c +/+	High carpellody of Class 1 or 2
2	c/c c/+ s/s	Partial carpellody of Class 2 or 3
2	c/+ c/c s/s	Partial carpellody of Class 2 or 3
1	c/c +/+ s/s	Partial carpellody of Class 3 or 4
2	c/c +/+ s/+	Partial carpellody of Class 3
1	c/c +/+ +/+	Partial carpellody of Class 3, possibly a pentandria type
4	c/+ c/+ +/+	Partial carpellody of Class 3 or 4
1	+/+ c/c s/s	Partial carpellody of Class 3 or 4
2	+/+ c/c s/+	Partial carpellody of Class 3
1	+/+ c/c +/+	Partial carpellody of Class 3, possibly a pentandria type
2	c/+ +/+ +/+	Normal or near normal type of Class 5 or 6
2	+/+ c/+ +/+	Normal or near normal type of Class 5 or 6
1	+/+ +/+ +/+	Normal type, used as normal parental type
2	+/+ +/+ s/+	Normal or near normal type of Class 6 or 7
4	c/+ +/+ s/+	Normal type, but not as stable as parental type, Class 5 or 6
4	+/+ c/+ s/+	Normal type, but not as stable as parental type, Class 5 or 6
8	c/+ c/+ s/+	Normal type, but not as stable as parental type, Class 5 or 6
4	c/+ c/+ s/s	Partially sterile type of Class 8 or 9
2	c/+ +/+ s/s	Partially sterile type of Class 8 or 9
2	+/+ c/+ s/s	Partially sterile type of Class 8 or 9
1	+/+ +/+ s/s	High sterile type of Class 10 or 11, used as female sterile parental type

[a]When either c locus is homozygous for c, carpellody is expressed regardless of phase of s locus. If no c locus is homozygous for c, s/s is sterile, s/+ is normal, +/+ is normal or slightly carpellodic depending on whether 0, 1, or 2 c alleles are present.

TABLE XII. OBSERVED AND CALCULATED RATIOS FOR PROGENIES OF CROSSES BETWEEN HIGH CARPELLODY, HIGH STERILE, AND NORMAL LINES

GENERATION	CROSS								
	HIGH CARPELLODY X NORMAL			NORMAL X HIGH STERILE			HIGH CARPELLODY X HIGH STERILE		
	Observed	Phenotype	Calculated	Observed	Phenotype	Calculated	Observed	Phenotype	Calculated
P_1		High Carpellody c/c c/c +/+			Normal +/+ +/+ +/+			High Carpellody c/c c/c +/+	
P_2		Normal +/+ +/+ +/+			High Sterile +/+ +/+ s/s			High Sterile +/+ +/+ s/s	
F_1		Partial Carpellody c/+ c/+ +/+			Normal +/+ +/+ s/+			Normal c/+ c/+ s/+	
F_2	11	High Carpellody c/c c/c +/+ c/c c/+ +/+	12	28	Normal +/+ +/+ +/+ +/+ +/+ s/+	30.75	12 11 11	High Carpellody Partial Carpellody Normal	7 11 12.5
	14	Partial Carpellody c/+ c/+ +/+	15				2	Partial Sterility	5
				13	High Sterile +/+ +/+ s/s	10.25	1	High Sterile	1.5
		X^2 = .48, p = .50			X^2 = 1.20, p = .25			(see Table XI for genotypes) X^2 = 5.72, p = .14	
BCP_1	Plants not available in this cross			Plants not available in this cross			6 12 7	High Carpellody Partial Carpellody Normal	12.5 9.375 3.125
								(see Table XI for genotypes) X^2 = 8.53, p = .01	
BCP_2	9	Partial Carpellody c/+ c/+ +/+	9.75	20	Normal +/+ +/+ s/+	17.5			
	30	Normal c/+ +/+ +/+ +/+ +/+ +/+	29.25	15	High Sterile +/+ +/+ s/s	17.5	Plants not available in this cross		
		X^2 = .059, p = .95			X^2 = .70, p = .40				

SUMMARY AND CONCLUSION

This study was conducted to determine the effects of temperature and rainfall on female sterility and carpellody of stamens and their inheritance patterns.

Histological examinations established that the sexual expression of the hermpahroditic flower was determined primarily by temperature variations some 6 to 8 weeks prior to anthesis. It was at that time prior to anthesis that the differentiation of organs occurred. It was also shown that breeding programs can develop lines of hermaphroditic papaya which are highly stable within certain limits. These lines and their crosses were then used to examine the temperature effects on sexuality and to determine the genetic pattern of inheritance of the factors for carpellody and sterility.

Basically, it was found that temperature effects were of an additive nature after reaching a basic threshold of sensitivity to temperatures inherent in the plant's genotype (of modifying factors). In most cases, the rate of sexual shift was about 1 magnitude of shift for each 1° to 3°F. of temperature change. Generally, the greater the amount of inbreeding, the higher the threshold value and the greater the temperature change required to overcome the threshold value.

Histograms of the parental lines and their crosses indicated a slight apparent dominance for the carpellodic factor and supported the theory of multiple genetic factors. The actual ratios of progenies obtained in the field can be closely approximated by using a tri-hybrid genetic model with "c", "c", and "s", and their normal allele (+), with

epistasis of "s" over "c" unless the "c" factor is homozygous, partial dominance of "+" over "c", and complete dominance of "+" over "s".

It is felt that crosses between the pentandria type and all other types, when combined with the data obtained in this study would be helpful in resolving the relationship between carpellody and sterility. It is also indicated that breeding for hermaphroditic lines of papaya which would exhibit sexual stability under a wide range of climatic conditions may not be easily attainable and that sexually stable cultivars may have to be developed solely for and within specific localities.

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